7th International Symposium on the Mediterranean Pig

Edited by
E.J. De Pedro and A.B. Cabezas

OPTIONS
méditerranéennes

SERIES A: Mediterranean Seminars
2012 – Number 101

CIHEAM

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7th International Symposium on the Mediterranean Pig

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Proceedings of the 7th International Symposium on the Mediterranean Pig, organised by the Escuela Técnica Superior de Ingeniería Agronómica y de Montes of the University of Córdoba
Córdoba (Spain), 14-16 October 2010

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Head of publication: Francisco Mombiela

2012 | Series A: Mediterranean Seminars | Number 101

Centre International de Hautes Etudes Agronomiques Méditerranéennes
International Centre for Advanced Mediterranean Agronomic Studies

L'édition technique, la maquette et la mise en page de ce numéro d'Options Méditerranéennes ont été réalisées par l'Atelier d'Édition de l'IAM de Zaragoza (CIHEAM)

Technical editing, layout and formatting of this edition of Options Méditerranéennes was carried out by the Editorial Board of MAI Zaragoza (CIHEAM)

Crédits des photos de couverture / *Cover photo credits*:
R. Cabrero

Tirage / *Copy number*: 350 ex.
Printer: INO Reproducciones, S.A.
Pol. Malpica, calle E, 32-39
(INBISA II, Nave 35)
50016 Zaragoza-Spain
Dep. Legal: Z-2893-91

Fiche bibliographique / *Cataloguing data*:

7th International Symposium on the Mediterranean Pig. E.J. De Pedro and A.B. Cabezas (eds). Zaragoza: CIHEAM / UCO, 2012, 677 p. (*Options Méditerranéennes*, Series A: Mediterranean Seminars, no. 101)

Catalogue des numéros d'Options Méditerranéennes sur /
Catalogue of Options Méditerranéennes issues on:
www.ciheam.org/publications

ISSN: 1016-121-X – ISBN: 2-85352-488-4

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Prólogo

En nombre del Comité Organizador y del Comité Científico, es un honor para mí presentar el trabajo con los resultados del 7º Simposio Internacional sobre el Cerdo Mediterráneo celebrado en Córdoba, del 14 al 16 de Octubre de 2010.

Han pasado más de 20 años desde que en 1989 en Ajaccio (Córcega) se celebró la primera edición. El tiempo transcurrido no ha debilitado la ilusión con la que un grupo de investigadores de España, Italia y Portugal, liderados por François Casabianca, actual director del Laboratorio de Investigación y Desarrollo de la Ganadería del INRA de Corte (Córcega), se propusieron compartir, debatir y difundir los avances que se dan en la producción porcina ligada a sistemas agrosilvopastorales. Prueba de ello son los Simposios precedentes a éste y la previsión de celebración de los próximos, ya acordados, en 2013 y 2016, así como la participación de investigadores de otras regiones europeas y del resto del mundo.

El camino andado no ha sido fácil, dado que es un sistema de producción con muy poco “peso específico” en la producción nacional y, en ocasiones, apenas tenido en consideración, sin olvidar las amenazas propias del sistema (la problemática de la base arbórea, sobreexplotación ganadera, epizootias, exigencias de calidad y seguridad alimentaria, crisis financiera, ….). Pero eso no nos ha hecho desistir de nuestro propósito de apostar por este tipo de encuentros, en los cuales las experiencias, avances, logros y fracasos, son compartidos con toda la comunidad científica, técnica y profesional, como si de una red de vasos comunicantes se tratara.

El objetivo es claro: lograr que este sistema de producción progrese, aportando a la sociedad todos los valores que posee: ser un elemento clave del desarrollo económico de regiones y pueblos, desempeñar un papel fundamental en la gestión medioambiental en zonas rurales asociadas a los entornos agrosilvopastorales y permitir una oferta de productos de alta calidad, muy deseados por otros tipos de producciones ganaderas.

Hoy día, en un mundo globalizado, debemos trabajar juntos, porque con el intercambio de estudios y experiencias salimos todos ganando: los que tienen ciertos problemas superados permitirán avanzar más rápidamente a aquellos operadores que están tratando de superar esos problemas, o aquellos que, no teniendo situaciones de crisis, pueden ayudar a tomar decisiones preventivas en aquellos lugares que las tienen o les pueden sobrevenir. En todo ello subyace una cultura: la de compartir, para lograr avanzar juntos, cada uno en la medida de sus posibilidades y de acuerdo con su entorno socioeconómico.

El sistema de producción porcina debe estudiarse en todo su contexto y, por tanto, en el Simposio se han tratado, como viene siendo habitual, cuestiones de genética, manejo productivo, nutrición, sanidad, productos y aspectos socioeconómicos. Dado que la calidad de los productos juega una baza importante en la supervivencia de estos sistemas, el 40% de los trabajos han tratado este aspecto, así como de la trazabilidad de los mismos (12%), con el fin de aportar transparencia en las transacciones comerciales y garantía a los consumidores en los diferentes escalones del sistema (ganadero – industrial – consumidor).

Con motivo de celebrar por segunda vez el Simposio en España, no puedo terminar sin un recuerdo para D. Armando Fallola, quien tuvo el honor de celebrar el 2º Simposio sobre el Cerdo Mediterráneo en Badajoz, en el año 1992, que falleció recientemente y con el que numerosos participantes a estos encuentros tenían una estrecha relación profesional, científica y técnica.

Para concluir, quisiera destacar el considerable trabajo conjunto llevado a cabo por los Comités Organizador y Científico, que ha hecho posible la realización del 7º Simposio sobre el Cerdo Mediterráneo. Aunque la situación económica existente en las fechas de celebración del evento era preocupante, debo manifestar mi agradecimiento a las entidades públicas y privadas, nacionales e internacionales que han colaborado con la organización del mismo, dando oportunidad a los participantes de compartir estos tres intensos días de trabajo y, por descontado, debo dar las gracias también a todos los participantes.

Emiliano de Pedro Sanz

Presidente del Comité Organizador y del Comité Científico

Foreword

On behalf of the Organizing and Scientific Committees, it is an honour for me to present the work with the results of the 7th International Symposium on the Mediterranean Pig held in Cordoba from 14 to 16 October 2010.

More than twenty years have passed since the first symposium was organized in 1989 in Ajaccio (Corsica). This long period of time has not weakened the enthusiasm with which a group of researchers from Spain, Italy and Portugal, led by Francois Casabianca, the current director of the Laboratory for Research and Development of Livestock in INRA Corte (Corsica), aimed to share, discuss and spread the progress achieved in pork production linked to sylvopastoral and agroforestry systems. Proof of this is the previous symposia and the plans to organize many more to come, in 2013 and 2016, as well as the participation of researchers from other European regions and the rest of the world.

The road travelled has not been easy, bearing in mind that this production system has very little "specific weight" in the whole national production sector and, on occasions, is even hardly taken into consideration, without forgetting the threats hovering over the system itself (the problem of the arboreal base, overexploitation of the livestock, epizootics, feed safety and quality requirements, financial crisis, etc.). But this has not prevented us from pursuing our goal to hold this type of meetings where experiences, advances, success and failure are shared by the scientific, technical and professional community, as though it were a network of communicating vessels.

The objective is clear: the advance of this production system, contributing all the values it possesses to society: being a key element in the economic development of regions and villages, playing a fundamental role in environmental management in rural zones associated with sylvopasture agroforestry areas and allowing the offer of high quality products much wanted in other types of livestock production.

Today, in our globalised world, we must work together, as with the exchange of studies and experience we all come out winning. Those who have overcome certain problems will help those operators who are trying to overcome the same problems to advance quicker or those that, not finding themselves in a critical situation, can help to make preventative decisions in places which are in or are close to a crisis. Underlying all this there is a culture: that of sharing to achieve advances together, each one within the limits of their possibilities and in agreement with their socioeconomic environment.

The pork production system must be studied within its wider context and, as such, issues related to genetics, productive management, nutrition, health, product and socioeconomic aspects have been dealt with in the Symposium, as is becoming habitual. Given that the quality of the product plays an important role in the survival of these systems, 40% of the work has dealt with this aspect, as well as the traceability of the systems (12%), with the aim of contributing transparency to commercial transactions and giving a guarantee to the consumers in the different stages of the system (stockbreeder, industry, consumer).

On the occasion of this second symposium in Spain, I cannot end without mentioning Mr. Armando Fallola, who had the honour to organize the second Symposium of the Mediterranean Pig in Badajoz in 1992, and who passed away recently and with whom numerous participants who attend these meetings had a close scientific, professional and technical relationship.

To conclude, I would like to make a point of the considerable team work carried out by the Organizing and Scientific Committees which has made the organization of the 7th Symposium of the Mediterranean Pig possible. Although the existing economic situation during the days of the Symposium was worrying, I must show my gratitude to public and private entities, both national and international, that have collaborated in the organization of this event, giving the participants the opportunity to share these three intensive days of work. And, needless to say, I must also thank all the participants.

Emiliano de Pedro Sanz

President of the Organizing and Scientific Committees

Avant-propos

Au nom du Comité Organisateur et du Comité Scientifique, c'est un honneur pour moi de présenter le travail et les résultats du 7ème Symposium International sur le Porc Méditerranéen qui a eu lieu à Cordoue du 14 au 16 octobre 2010.

Plus de 20 ans ont passé depuis qu'en 1989 à Ajaccio (Corse) la première édition fut célébrée. Le déroulement du temps n'a pas affaibli l'intention avec laquelle un groupe de chercheurs espagnols, italiens et portugais, dirigés par François Casabianca, actuel directeur du Laboratoire de Recherche sur le Développement de L'Elevage de l'INRA de Corte (Corse), se sont proposés de partager, débattre et de diffuser les progrès se réalisant dans la production porcine liée aux systèmes agro-silvo-pastoraux. Les Symposiums précédents et la prévision de la tenue des prochains, déjà décidés pour 2013 et 2016, ainsi que la participation de chercheurs d'autres régions européennes et du reste du monde en sont la preuve.

Le chemin parcouru n'a pas été facile, étant donné qu'il s'agit d'un système de production doté de «peu de poids spécifique» dans la production nationale et, en d'autres occasions, tout juste pris en considération, sans oublier les menaces pesant sur le système lui-même (la problématique de la base arborée, les épizooties, les normes de qualité et de sécurité alimentaire, la crise financière…). Mais tout ceci ne nous a pas fait reculer devant notre projet de parier pour ce type de rencontres, au cours desquelles les expériences, progrès, satisfactions et échecs, sont partagés par toute la communauté scientifique, technique et professionnelle, comme s'il s'agissait d'un vaste réseau de vases communicants.

L'objectif est clair: faire en sorte que ce système de production progresse, apportant ainsi à la société, toutes les valeurs qu'il possède: être un élément clef du développement économique des régions et des villages, jouer un rôle fondamental dans la gestion de l'environnement dans les zones rurales associées aux environnements agro-sylvo-pastoraux et permettre d'offrir des produits de haute qualité, enviés d'autres types de productions d'élevage.

De nos jours, dans un monde globalisé, nous devons travailler ensemble, parce qu'en échangeant nos études et nos expériences, nous sommes tous gagnants : ceux qui ont trouvé solution à leurs problèmes permettront à ceux qui essaient de résoudre ces mêmes problèmes de progresser plus rapidement, ou bien ceux qui, sans être touchés par la crise, apportent leur aide pour prendre des décisions préventives dans les zones où elles existent ou bien elles peuvent survenir. Dans tout ceci une culture est sous-jacente: celle de partager, pour arriver à progresser ensemble, chacun dans la mesure de son possible et en accord avec son environnement socio-économique.

Le système de production porcine doit être étudié dans la globalité de son contexte et nous avons donc traité, comme d'habitude au cours du Symposium, des questions de génétique, la gestion de production, la nutrition, la santé, les produits et les aspects socio-économiques. La qualité des produits jouant un atout important dans la viabilité de ces systèmes, 40% des travaux concernents ces aspects, de même que leur traçabilité (12%), avec pour objectif d'apporter la transparence dans les transactions commerciales et la garantie aux consommateurs aux différents échelons du système (élevage, industriel, consommateur).

À l'occasion d'avoir célébré une deuxième fois le Symposium en Espagne, je ne peux terminer sans rendre hommage à M. Armando Fallola, récemment décédé, qui a eu l'honneur de présider le 2ème Symposium sur le Porc Méditerranéen à Badajoz en 1992, et avec qui de nombreux participants à ces rencontres maintenaient une étroite relation professionnelle, scientifique et technique.

Pour conclure, je voudrais souligner le travail considérable mené à bien par les Comités Organisateur et Scientifique, qui ont rendu possible la réalisation du 7ème Symposium sur le

Porc Méditerranéen. Malgré une situation économique inquiétante pendant la période de tenue de cette réunion, je voudrais remercier tous les organismes publics et privés, nationaux et internationaux ayant collaboré avec l'organisation, donnant l'opportunité aux participants de partager ces trois jours intenses de travail et je voudrais évidemment remercier tous les participants.

Emiliano de Pedro Sanz

Président des Comités Organisateur et Scientifique

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Session 1
Genetics

Sequencing the Iberian pig genomes

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Abstract. The Iberian pig is one of the most important varieties of local pig breeds, both economically and culturally. Contrary to most widespread breeds, like Duroc or Large White, it seems not to have been intercrossed with Chinese pigs. Modern sequencing technologies allow us to sequence and analyze complete genomes for costs in the order of a few thousand euros, democratizing the access to this kind of data. Here we present preliminary results of the partial sequencing a highly inbred animal of the Guadyerbas strain.

Keywords. Iberian pig – Guadyerbas strain – Next Generation Sequencing.

Séquençage des génomes du porc ibérique

Résumé. *Le porc Ibérique est une des plus importantes races locales, économiquement et culturellement. Au contraire d'autres races comme Duroc et Large White, il n'est pas hybridé avec des races asiatiques. Les techniques de séquençage modernes nous permettent de séquencer et d'analyser des génomes complets pour un prix raisonnable, démocratisant les outils génomiques. On présente ici les résultats préliminaires de la séquence d'un porc Ibérique de la lignée Guadyerbas.*

Mots-clés. *Porc Ibérique – Guadyerbas – Séquençage à haute performance.*

I – Introduction

Next generation sequencing (NGS) has revolutionized genomics research, making it difficult to overstate its impact in Biology. NGS will immediately allow researchers working in non mainstream species to obtain complete genomes together with a comprehensive catalogue of variants. A reasonable question to ask, nevertheless, is whether we *really* need so much more sequence. Our answer is yes, we do. There are important advantages on having full sequence rather than genotypes: removing SNP ascertainment bias, uncovering all extant variability, recovering the full unbiased demographic history of populations, or setting the theoretical ground for a unified framework that combines coalescence and association mapping. Additional applications involve RNA-seq, to quantify precisely the transcriptome and study allele specific expression.

We have been studying the Iberian pig from a genetic point of view for many years. The Iberian pig is one of the most important varieties of local pig breeds, both economically and culturally. Contrary to most widespread breeds, like Duroc or Large White, it seems not to have been intercrossed with Chinese pigs. However, a current danger for Iberian germplasm is uncontrolled introgression with Duroc genomes, a fact that may have been exacerbated by recent increasing demand of Iberian pig products. Here, we present ongoing efforts carried out at the Universitat Autonoma de Barcelona and INIA in cooperation with the Centre for Genomics Research in Barcelona, University of Vienna, and Wageningen University in Holland to sequence several Iberian pig genomes and transcriptomes. Thus far, we only have analyzed about 1% of the Guadyerbas strain, and this is what is discussed in this communication.

II – Methods

The Guadyerbas herd was founded with four boars and ten sows in 1945; and has been maintained with controlled pedigree and minimum co-ancestry mating practices in order to minimize increase in inbreeding (Odriozola, 1976). Despite this, and because of isolation and small number of breeding animals, average inbreeding coefficient F is very high for all surviving pigs. In the specific female sequenced, autosomal F was ~ 0.40 and ~ 0.46 for X chromosome.

We prepared a Reduced representation Library (RRL) by digesting with HaeIII enzyme and selecting the band of 200 ± 10 bp. From the three runs, a total of 25.3 million base called reads were obtained. Reads were trimmed to 40 bp due to low 3' end quality. We aligned the quality filtered sequences against the reference porcine genome assembly 9 (ftp://ftp.sanger.ac.uk/pub/S_scrofa/assemblies/Ensembl_Sscrofa9/) with GEM (http://sourceforge.net/apps/mediawiki/gemlibrary/index.php?title=The_GEM_library), MAQ (Li *et al*., 2008) and Mosaik (http://bioinformatics.bc.edu/marthlab/Mosaik) allowing up to 3 mismatches and we retained for variant calling only those reads that mapped unambiguously. We identified SNPs with GEM, MAQ and GigaBayes (Quinlan *et al*., 2008). When mapping the filtered reads with GEM, we used default options except for the mismatches allowed in each read to the reference genome. For the identification of SNP we used custom Perl scripts. For the assignation of SNP_F, the minimum allele frequency of the alternative allele was set to 0.9. For SNP_H with a coverage of 3×, the reference allele had to be found 1× and the alternative allele 2×; for the rest of the cases ($SNP_H \geq 4\times$), the minimum allele count was 2, the maximum frequency of the reference allele was 0.4 and the maximum frequency of any allele was 0.8%.

After filtering, see methods, and removing ambiguous matching reads, we retained five million reads for further analysis. The total length assembled was 2.3 Mb. The reads spanned 83.1 Mb of the porcine assembly v. 9 with at least one read, and 25.1 Mb with at least three reads. The average coverage, counting only regions with read depth between 3 and 20 was 4×. All chromosomes were uniformly covered and we did not notice biases regarding read distribution within chromosomes.

We classified the SNPs into two classes, fixed (SNP_F) when the differences were between the assembly and the Iberian reads, and segregating (SNP_H) when the Iberian pig was heterozygous. We computed the average number of SNPs, fixed differences and heterozygous, along non - overlapping contiguous 500 kb windows. We also obtained Hudson – Kreitman – Aguadé (HKA) diversity (θ) estimates (Hudson *et al*., 1987). Briefly, HKA tests whether there is a deviation between observed and expected number of polymorphisms, where the expected polymorphism is obtained from the divergence between an outgroup and the population studied.

III – Results and discussion

We found a global autosomal Iberian heterozygosity rate of $\pi = 0.5\times10^{-3}$. This value should be taken with some caution, though, because it might be inflated by false positives; alternatively, some true SNPs have been certainly discarded because of stringent criteria in identifying SNPs. Nevertheless, assuming a mutation rate (μ) of 10^{-8}, this results in an estimate of effective size $Ne = \pi/4\mu \sim 10^4$. This value is larger than expected, in particular considering that this is a highly inbred animal and suggests that the actual effective size in the founder herd might be actually double. When correcting for inbreeding, this diversity is comparable to that reported in other porcine species (Amaral *et al*., 2009a; Amaral *et al*., 2009b) or in humans. Therefore, we can hypothesize that loss of genetic variability in the Iberian pig population is very recent and caused by recent inbreeding. Sequencing more animals is needed to investigate these issues further.

To gain further insight into the variability distribution, we plotted the percentage of fixed and segregating SNPs in non-overlapping contiguous windows of 500 kb, Figure 1 shows the results for two chromosomes, SSC1 and SSCX as examples. For clarity, we also present the lowess adjusted curves. A trend of increasing variability toward the telomeres is clearly visible, both in fixed and in heterozygous SNPs. This occurs as well in the sex chromosome, although less marked than in autosomes because the overall level of variability is lower.

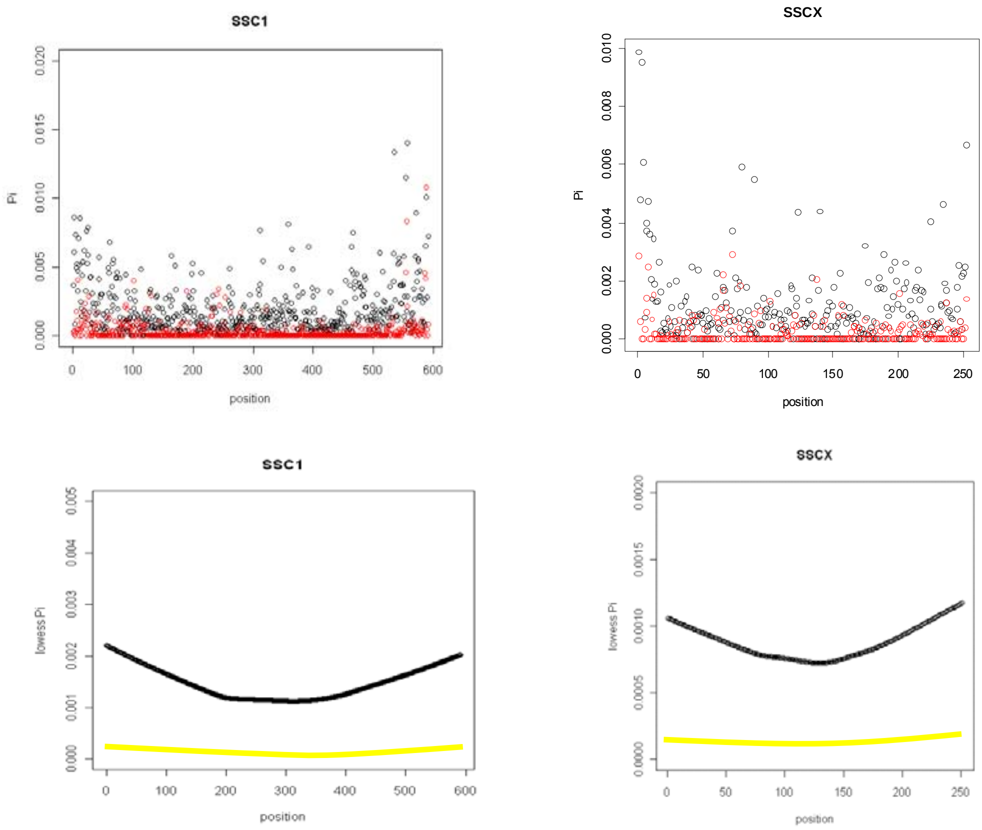

Fig. 1. Average rate of fixed (dark) and heterozygous (light) SNPs per bp in 500 kb windows (Pi) along chromosomes 1 and X. Position refers to window number. The top panels show the observed values whereas the bottom panels are the lowess adjusted curves, shown to underline an increased variability towards the telomeric regions.

Overall, the HKA test showed no strong departures of neutrality. Certainly, not the whole genome evolves according to the standard neutral model and the apparent neutrality may simply mean lack of power or too large windows that may mask highly local selective events.

IV – Conclusion and perspectives

Having complete genomes sequenced is no longer a luxury available only to large sequencing centers. Our challenge now is to convert this huge amount of information into useful knowledge for the conservation and improvement of local breeds, as well as to identify the variants responsible for the distinctiveness of these breeds compared to highly selected, international breeds. On our side, the next immediate steps are: i) to sequence the transcriptome in a few Iberian and Large White samples; ii) to sequence pools of Iberian pigs; iii) to increase coverage of the Guadyerbas animal sequenced; and iv) to sequence partially American pigs of Iberian descent adapted to extreme climate environments, i.e., altitude and heat.

Acknowledgments

Conservation of the *Guadyerbas* strain started with late Miguel Odriozola and has been ensured for years thanks to the efforts of L. Silió, J. Rodrigáñez and colleagues, and supported economically by the regional government of *Junta de Castilla-La Mancha* and *Instituto Nacional de Investigación y Tecnología Agraria y Alimentaria* (INIA). AE is funded by the Spanish Ministry of Research PhD studentship program (FPI), RK was supported by GABI-FUTURE grant BeetSeq (FKZ 0315069A) of the German Federal Ministry of Education and Research, work funded by grants AGL2007-65563-C02/GAN to MPE and MCR, and by a Consolider grant from Spanish Ministry of Research.

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Analysis of the *KIT* gene in a Sicilian pig population and identification of the I^d allele at the *Dominant white* locus

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Abstract. Mutations in the porcine *KIT* gene (*Dominant white* locus) have been shown to affect coat colors and color distribution in pigs. Classical genetic studies have indicated that the gray/roan coat colour observed in different pigs is determined by an allele (I^d) at the *Dominant white* locus. In a survey of the pig populations of the Nebrodi mountain area (Sicily), we identified a few animals showing morphological characteristics resembling the Nero Siciliano genetic type but with gray/roan (determined by a close intermingling of white and black hairs) instead of black hair, having, in several cases, depigmented skin areas. According to the data we obtained on crosses between gray/roan and black pigs, the mode of inheritance of the roan phenotype is due to a dominant allele over black. All 21 exons and intronic regions of the *KIT* gene were sequenced in gray/roan and in several other pigs. The gray/roan haplotype carried a 4 bp deletion in intron 18 but did not include any duplication of the *KIT* gene. Analysing polymorphic site co-segregation with the gray/roan phenotype provided evidence that this *KIT* allele may identify the I^d allele described in the early genetic literature.

Keywords. Coat colour – *Dominant white* locus – KIT – DNA polymorphisms – Roan.

Analyse du gène KIT et identification de l'allèle I^d du locus blanc dominant au sein de la population porcine en Sicile

***Résumé**. Les mutations au niveau du gène KIT (locus blanc dominant) affectent la couleur de la robe et la répartition des pigments chez le porc. Les études de la génétique classique suggèrent que le phénotype gris/rouan de la robe chez le porc est déterminé par un allèle (I^d) du locus blanc dominant. Lors d'une évaluation de ce patron de coloration au sein de populations porcines dans les régions montagneuses de Nebrodi (Sicile) nous avons observé que certains animaux présentant des caractéristiques morphologiques similaires à ceux de la race Nero Siciliano possèdent une robe grise/rouanne (mixture de poils blancs et noirs) plutôt qu'une robe noire. Par ailleurs, certains d'entre eux présentent des régions cutanées dépigmentées. Le croisement entre des porcs gris/rouans et des porcs noirs révèle que l'allèle rouan est dominant sur l'allèle noir. Nous avons séquencé les 21 exons et les régions introniques du gène KIT chez plusieurs animaux gris/rouans mais aussi chez d'autres animaux porcins. Les animaux de phénotype gris/rouan présentent une délétion de 4 pb au niveau de l'intron 18 sans aucune duplication du gène KIT. Cet allèle du gène KIT co-ségrège avec le phénotype gris/rouan et, par conséquent, correspondrait à l'allèle I^d précédemment décrit par l'analyse génétique.*

***Mots-clés.** Couleur de la robe – Locus* Dominant white *– KIT – Polymorphismes de l'ADN – Rouan.*

I – Introduction

Several studies on coat colour genetics in pigs have identified that mutations in the melanocortin 1 receptor (*MC1R*) and v-kit Hardy-Zuckerman 4 feline sarcoma viral oncogene homolog (*KIT*) genes are the major determinants of the variability of this phenotypic trait among and within different pig breeds (Kijas *et al.*, 1998; Marklund *et al.*, 1998; Giuffra *et al.*, 1999;

Kijas *et al.*, 2001; Johansson *et al.*, 2005; Fang *et al.*, 2009; Fontanesi *et al.*, 2010a). Epistatic interactions between these two loci have been reported in QTL analysis for this trait (Hirooka *et al.*, 2002). The *KIT* gene encodes the mast/stem cell growth factor receptor that is involved in driving the melanocyte migration from the neural crest along the dorsolateral pathway to colonize the final destination in the skin (Besmer *et al.*, 1993).

A large number of *KIT* mutations have been reported to cause pigmentation anomalies in several species (i.e. Jackson, 1997). In cattle, selection signature at the *KIT* gene is evident in a few breeds in which polymorphisms are associated with the spotting phenotype (Fontanesi *et al.*, *2*010b). In pigs, the *KIT* gene maps on chromosome 8 and causes the *Dominant white* (*I*) locus (Johansson *et al.*, 1992). An unusual large number of alleles, each with different or partially overlapping phenotypic effects, have been characterized at this locus (Johansson Moller *et al.* 1996; Marklund *et al.*, 1998; Giuffra *et al.*, 1999; Giuffra *et al.*, 2002; Pielberg *et al.*, 2002; Johansson *et al.*, 2005): the recessive wild type allele (*i*) is usually associated with a solid or wild type coat color; the patch allele (I^P) causes the presence of colored patches on a white background (Johansson Moller *et al.*, 1996); the *Belt* allele (I^{Be}) determines the white belt observed in the Hampshire and Cinta Senese breeds (Giuffra *et al.*, 1999; Fontanesi *et al.*, 2010a); four *Dominant white* alleles (I^1, I^2, I^3 and I^L) determine white uniform colored pigs, even if in few cases black spots have been reported in animals carrying some of these forms (Pielberg *et al.*, 2002; Johansson *et al.*, 2005). A few of these alleles (*i*, I^{Be} and I^L) have a single copy of the *KIT* gene whereas all other alleles are determined by copy number variation (CNV) of this gene with a duplication or triplication of a region including the whole coding sequence, encompassing 21 exons, and flanking regions (Giuffra *et al.*, 2002; Pielberg *et al.*, 2002). In addition to the gene duplication, the I^1 allele carries a splice mutation in one of the two copies at the level of intron 17 (causing the skipping of exon 17, predicted to impair an important function of the coded protein) in linkage disequilibrium with a 4 bp deletion in intron 18 (*intron18-g.29_32delAGTT*) (Marklund *et al.*, 1998; Giuffra *et al.*, 2002). In the I^2 and I^3 alleles the splice mutation is in one or two copies of the three-copies gene forms, respectively (Pielberg *et al.*, 2002).

It seems possible that other alleles with a larger number of copies and combinations with the splice mutation are present in white pig populations (Johansson *et al.* 2005; our unpublished data). The I^P allele lacks the splice mutation present in the other duplicated alleles (Johansson Moller *et al.*, 1996). The I^L allele contains a single mutated copy of the *KIT* gene (with the splice mutation) and might be lethal if homozygous (Pielberg *et al.*, 2002; Johansson *et al.*, 2005). The I^{Be} allele is probably caused by a not yet characterized regulatory mutation (Giuffra *et al.*, 1999). In addition to this complex allele series, earlier studies on pig coat color indicated the presence of an additional allele (I^d), not characterized yet at the molecular level, giving a gray-roan phenotype and dominant over the *i* allele (Hetzer, 1948; Lauvergne and Canope, 1979).

Nero Siciliano breed (Sicilian Black) is reared mainly under extensive management in the Nebrodi and Madonie mountains of the provinces of Messina and Palermo on the island of Sicily. This is a local population whose genetic pool has been shaped by subsequent introduction of blood from other local or cosmopolitan populations and breeds. In addition to the completely black animals, Nero Siciliano population includes pigs with white portions, mainly in the head and legs. Recently, in a survey of the pig populations of the Nebrodi mountain, we identified several animals showing morphological characteristics resembling the Nero Siciliano genetic type but with gray/roan instead of black hair, having almost always some pigmented skin areas and dark/black hair in the head. This coat color phenotype resembles the gray/roan coat color described in the early literature (McLean, 1914; Hetzer, 1948; Lauvergne and Canope, 1979), assumed to be caused by the I^d allele at the *Dominant white* locus.

In this study we characterized by sequencing and/or genotyping the *KIT* and *MC1R* genes of these pigs, compared these data with those we obtained for other breeds and provided evidences that the *KIT* allele in the gray/roan animals should correspond to the I^d allele.

II – Materials and methods

Animals investigated in this study were 42 gray/roan pigs sampled from 2 different farms in Sicily (14 and 28 respectively) (D'Alessandro *et al.*, 2007; Fontanesi *et al.*, 2010). Twenty-one of these gray pigs, derived from one farm, were obtained by crossing 7 Nero Siciliano black sows with one gray boar. Eighteen completely black pigs were obtained from the same litters. For the other gray pigs pedigree information was not available. Of the 42 gray hair pigs, almost all (38) presented pigmented skin areas of variable extension. Blood or hair root samples were collected from these pigs and DNA was extracted using standard protocols. DNA samples were also available for other pigs used for sequencing (see below) (D'Alessandro *et al.*, 2007; Fontanesi *et al.*, 2010).

Mutations in the *MC1R* and *KIT* genes were analysed by PCR-RFLP or amplified fragment analysis as previously reported (Kijas *et al.*, 1998; Marklund *et al.*, 1998; Giuffra *et al.*, 2002; Kijas *et al.*, 2002; D'Alessandro *et al.*, 2007; Fontanesi *et al.*, 2010a).

The complete coding region (spanning 21 exons) and partial upstream and downstream intronic regions of the *KIT* gene was sequenced in pigs of different breeds and coat colour (Fontanesi *et al.*, 2010a): 10 Nero Siciliano pigs, 4 with completely black coat color, 4 with white spots but without duplication of the *KIT* gene, 2 with white spots but with the duplication of the *KIT* gene; 3 gray animals (one was homozygous for intron 18 deletion and two were heterozygous at this polymorphic site); 6 Cinta Senese; 4 Hampshire; 4 Duroc, 2 Italian Large White, one carrying the splice mutation and one not carrying this mutation; 2 Pietrain, one positive and one negative at the duplication test; 2 Meishan; 2 wild boars. Polymorphisms were detected using CodonCode Aligner (http://www.codoncode.com/aligner) with the AC141857 sequence (pig genomic sequence derived from clone RP44-473N18 containing the 21 *KIT* exons) included as a reference (Fontanesi *et al.*, 2010a).

Calculation of the *LOD* score in the half sibling family structure was obtained with the LODS program (Linkage Utility Programs, Rockefeller University). Polymorphisms determined by sequencing of the *KIT* gene were used to reconstruct haplotypes with PHASE program v. 2.1.1 (Stephens *et al.*, 2001). Phylogenetic analyses of the identified haplotypes of the *KIT* gene were conducted with MEGA4 software v. 4.0.2 (Tamura *et al.*, 2007) with Kimura's two parameter model and Neighbour Joining (NJ) trees using sequence information and polymorphisms identified in the coding regions including also other porcine *KIT* cDNA sequences available in GenBank database. Standard errors were obtained from 1000 bootstrap replicates.

III – Results and discussion

Genotyping the grey/roan pigs for two polymorphisms of the *KIT* gene (*intron18-g.29_32delAGTT* and duplication test; Johansson Moller *et al.*, 1996; Giuffra *et al.*, 2002) unusual results were obtained. Interestingly, all these animals carried at least one *KIT* allele with the deletion of 4 bp in intron 18 (*intron18-g.29_32delAGTT*), typical of one of the two (or three) *KIT* copies of the I^1, I^2 or I^3 alleles of the *Dominant white* locus. However, no duplication of the *KIT* gene was observed in these animals, that means CNV was not present in the *KIT* alleles of these pigs. Pedigree relationships recorded in one of the two farms demonstrated that 21 gray/roan pigs (heterozygous for the 4 bp deletion of intron 18) derived from 7 litters obtained crossing a gray/roan boar (heterozygous for the *KIT intron18-g.29_32delAGTT* allele) with 7 different black Nero Siciliano sows (that did not carry the 4 bp deletion of intron 18)

(Fontanesi *et al.*, 2010a). Eighteen other pigs of the same litters were completely black. These black piglets did not carry the 4 bp deleted allele. Considering all these litters, no deviation from the classical 1:1 Mendelian ratio of the two coat color phenotypes (gray/roan and black) was evident, suggesting dominance of the gray/roan phenotype (associated with the *KIT intron18-g.29_32delAGTT* allele) over the colored (wild type) *KIT* allele(s) of the black sows. Association between the *KIT intron18-g.29_32delAGTT* allele (in heterozygous condition) and gray coat color in these 7 litters was highly significant (θ = 0.00; *LOD* = 11.74). All these animals were also genotyped for mutations in the *MC1R* gene. Gray/roan piglets of these litters had several genotypes (E^{D2}/E^{D2}, E^{D2}/E^{P}, or E^{P}/E^{P}) indicating that the genotype at the *MC1R* locus did not affect coat color of the gray animals (skin areas were pigmented in all three genotypes). It is also worth to mention that the wild boar *MC1R* allele (E^{+}), that should give a wild gray coat color, was not observed in these animals (Fontanesi *et al.*, 2010a).

A total of 6.7 kb of the *KIT* gene, sequenced in 35 pigs from 9 breeds and populations, provided a total of 69 polymorphisms, including 65 single nucleotide polymorphisms (SNPs) and 4 indels (Fontanesi *et al.*, 2010a). These data were used for phylogenetic analysis of the reconstructed haplotypes obtained for the different sequenced pigs (data not shown). Results indicated that the gray/roan haplotype was very close to the wild boar haplotype, suggesting that it might have been originated from a wild type sequence.

IV – Conclusions

This study provided evidences that the *KIT intron18-g.29_32delAGTT* mutation (but in a single copy *KIT* gene) characterizes the I^{d} allele at the *Dominant white* locus that was already described in the early pig coat color genetic literature. Considering the structure of the gray/roan *KIT* haplotype and that of the other *Dominant white KIT* alleles, it could be possible to reconstruct the mutational events that contributed to create the large heterogeneity observed at the *Dominant white* locus. The I^{d} allele might be one of the two copies of the I^{P} allele that was originated from a first ancestral duplication at this locus. Then, another mutational event at the level of intron 17 (splice site mutation) occurring at this putative I^{d} form in a duplicated allele could have created the I^{1} allele. Subsequent unequal crossing over events might have originated other alleles with more that two duplicated *KIT* genes. The example of the pig *Dominant white* locus could indicate that CNVs might be important source of genetic variability in livestock affecting phenotypic and , possibly, production traits.

Acknowledgments

This work was supported by MiPAAF Selmol (LF), University of Bologna RFO (LF, VR) and Regione Sicilia (VC) funds.

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Genetic characterization of autochthonous pig breeds from Spain with microsatellite markers

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Abstract. The European pig genetic resources are essentially formed from breeds belonging to two ancient strains: the Celtic type in north-central Europe and the Mediterranean type in the south. Both types were represented in Spain by native breeds, but today only the Iberian pig is completely developed. Other autochthonous breeds are rare, endangered or extinct. Population structure and genetic diversity in the Spanish native breeds of pigs Ibérico (ten varieties), Chato Murciano, Negro Canario, Negro de Formentera e Ibiza, Celta and Euskal Txerria were analyzed by typing 25 microsatellite markers recommended by the Food and Agriculture Organization of the United Nations. Within and between breed genetic variation was estimated by heterozygosities, F-statistics estimate, genetic distances and assignment tests. The results of this study confirm that Spanish local breeds of pigs represent a very interesting reservoir of allelic diversity, even though the current levels of inbreeding observed in some of the studied breeds. Therefore, appropriate conservation efforts should be undertaken, such as adopting strategies aimed at minimizing inbreeding, to avoid further losses of genetic diversity.

Keywords. Spanish local pigs – Genetic relationships – Conservation – Genetic structure.

Caractérisation des races autochtones porcines espagnoles avec des marqueurs microsatellitaires

Résumé. *La plupart des variétés européennes sont principalement de deux souches: le type Celte du Nord et d'Europe centrale et le type du sud de la Méditerranée. Les deux types sont représentés en Espagne par des races de porcs autochtones, et bien que certaines variétés de porcs ibériques soient pleinement mises en œuvre, d'autres variétés et la plupart des autres races sont fortement menacées. Nous avons étudié la structure et la diversité génétique des races de porcs espagnoles : Ibérico (une dizaine de variétés), Chato Murciano, Negro Canario, Negro Mallorquin, Negro de Formentera et Ibiza, Celta et Txerria Euskal ont été analysés avec 24 microsatellites recommandés par la FAO. La variation génétique au sein des races et entre les races a été étudiée par analyse d'hétérozygotie, par les indices de fixation, les distances génétiques et les études génétiques et l'affectation des individus aux populations. Les résultats confirment que ces animaux sont un sujet très intéressant de diversité génétique, en dépit des niveaux élevés de consanguinité observés dans certaines races. Cette étude est une première étape qui fournit des informations génétiques sur l'état de ces importantes ressources génétiques et en tenant compte de cette information, les programmes de conservation doivent être mis en œuvre dans certaines races pour éviter de nouvelles pertes de diversité génétique.*

Mots-clés. *Races porcines espagnoles – Relations génétiques – Conservation – Structure génétique.*

I – Introduction

Until the 1950s, swine production in Southern Europe was essentially based on native breeds raised under extensive systems, often in specific ecosystems integrating forest lands (Vicente and Alés, 2006). The intensification of agriculture that took place after that time caused major changes to pig breeding, with traditional systems being replaced by intensive production based on a reduced number of exotic breeds, while native breeds were progressively abandoned and became virtually extinct (Gama, 2006). The European pig genetic resources are essentially formed from breeds belonging to two ancient strains: the Celtic type in north-central Europe and the Mediterranean type in the south (Porter and Tebbit, 1993). Both types were represented in Spain by native breeds, but today only some varieties of the Iberian pig are completely developed (Retinto and Entrepelado). Other autochthonous breeds are rare, endangered or extinct. Currently, 7 native breeds of pigs are recognized in Spain (i.e., Ibérico, Chato Murciano, Celta, Negro Canario, Negro Mallorquín, EuskalTxerria and Gochu Asturcelta).

Detailed knowledge of population structure among and within breeds of livestock is essential for establishing conservation priorities and strategies (Caballero and Toro, 2002). Microsatellite markers have proved extremely useful for the analysis of population structure and relationships, and have been widely used for genetic characterization of several species and populations including European pig breeds (Laval *et al.*, 2000; Martínez *et al.*, 2000; San Cristobal *et al.*, 2006; Vicente *et al.*, 2008). Nevertheless, information on native breeds of pigs is still scarce, even though they possess unique characteristics in terms of adaptation, hardiness, and quality of products. In this study we used microsatellite markers i) to evaluate the degree and pattern of genetic variability in 6 native breed and one not officially recognised pig population currently used in Spain, and ii) to assess with different statistical tools the integrity and degree of admixture of these native swine breeds.

II – Materials and methods

Individual blood or hair samples were collected from 593 representative animals of the 15 populations under analysis. Specifically, the breeds studied and corresponding sample sizes are shown in Table 1.

DNA was extracted with Chelex 100 (Bio-Rad Laboratories, Alcobendas, Spain) and proteinase-K (Qbiogen, Illkirch, France), as described by Walsh *et al*. (1991). A panel of 24 microsatellite markers was established, according to the recommendations of the Food and Agriculture Organization of the United Nations and the International Society for Animal Genetics (FAO, 2004)*: IGF1, S0002, S0005, S0026, S0068, S0090, S0101, S0155, S0178, S0215, S0225, S0226, S0227, S0228, S0355, S0386, SW24, SW72, SW240, SW632, SW857, SW911, SW936 SW951.* Microsatellite markers were amplified in multiplex polymerase chain reactions (PCRs) using fluorescence-labelled primers according the methodology used by Vicente *et al*., 2008. PCR products were separated by electrophoresis on a ABI 377XL instrument (Applied Biosystems, Foster City, CA) according to manufacturer recommendations and allele sizing was accomplished by using the internal size standard GeneScan-400HD ROX (Applied Biosystems, Warrington, UK).

Results of electrophoresis were read directly and interpreted with GeneScan and Genotyper software (Applied Biosystems, Applera Europe B.V.), respectively. A first approach to within-breed diversity was investigated by calculating the mean number of alleles, observed and unbiased expected estimates of heterozygosity per population (Nei, 1987) and their standard deviations using "Microsatellite Toolkit" software (Park, 2001). F_{ST}, F_{IT} statistic and within-breed inbreeding coefficient (F_{IS}) (Weir and Cockerham, 1984) were estimated with a 95% confidence interval, determined by 10000 bootstraps across loci using "Genetix" v4.04 (Belkhir *et al.*, 2003). Genetic divergence among breeds was estimated through D_A genetic distances (Nei *et al.*, 1983), with the "Populations" software (Langella, 1999).

The "Structure" v.2.1 software (Pritchard *et al.*, 2000) was used to investigate the genetic structure of the 16 populations, in order to identify population substructure and admixture. Runs of 300000 iterations after a burn-in period of 100000 iterations were performed for each K, to determine the most probable number of clusters, as inferred from the observed genotypic data. The "Distruct" v.1.1 software (http://rosenberglab.bioinformatics.med.umich.edu/distruct.html) was used to obtain a graphical display of individual membership coefficients in each ancestral population.

Table 1. Number of samples analysed (N), mean number of alleles per locus (MNA), average expected heterozygosity (He), average observed heterozygosity (Ho) and their standard deviations, inbreeding coefficient (F_{IS}) with their confidence interval and number of loci with deviations from Hardy-Weinberg equilibrium (HWE) (p<0.05) estimated with 24 microsatellites in 16 Spanish pig populations

Population	N	Mna (sd)	He[1] (sd)	Ho (sd)	Fis (ic)	Hwed
Retinto portugués[2]	14	3.75 (1.62)	0.521 (0.046)	0.551 (0027)	-0.059 (-0.201 - -0.009)*	0
Retinto[2]	88	6.38 (2.81)	0.545 (0.047)	0.508 (0.011)	0.068 (0.025 - 0.099)*	7
Entrepelado[2]	73	6.21 (2.48)	0.564 (0.051)	0.543 0.012)	0.036 (-0.008 - 0.066)	7
Mamellado[2]	7	3.46 (1.69)	0.509 (0.058)	0.520 (0.039)	-0.023 (-0.300 - 0.007)	0
Silvela[2]	14	4.00 (1.56)	0.568 (0.047)	0.510 (0.028)	0.106 (-0.038 - 0.154)	1
Dorado gaditano[2]	7	3.29 (1.33)	0.495 (0.055)	0.500 (0.039)	-0.012 (-0.299 - 0.054)	1
Torbiscal[2]	60	5.00 (2.50)	0.527 (0.051)	0.465 (0.013)	0.118 (0.066 - 0.151)*	7
Negro de los Pedroches[2]	29	4.21 (1.41)	0.561 (0.040)	0.554 (0.020)	0.012 (-0.101 - 0.076)	2
Lampiño[2]	59	6.08 (2.99)	0.573 (0.048)	0.533 (0.013)	0.070 (0.012 - 0.111)*	6
Manchado de Jabugo[2]	41	3.38 (1.64)	0.388 (0.051)	0.397 (0.016)	-0.021 (-0.088 - 0.019)	3
Chato murciano	64	4.04 (1.27)	0.515 (0.042)	0.521 (0.013)	0.056 (0.011 - 0.090)*	14
Negro canario	27	3.96 (1.76)	0.474 (0.049)	0.475 (0.020)	-0.001 (-0.110 - 0.059)	1
Negro de Formentera e Ibiza	20	3.88 (1.62)	0.415 (0.044)	0.356 (0.022)	0.146 (-0.011 - 0.215)	6
Negro mallorquin	17	4.42 (1.50)	0.562 (0.032)	0.516 (0.025)	0.084 (-0.035 - 0.132)	4
Euskaltxerria	46	3.00 (1.47)	0.369 (0.052)	0.382 (0.015)	-0.034 (-0.092 - 0.002)	1
Celta	27	4.54 (1.79)	0.596 (0.047)	0.496 (0.021)	0.171 (0.079 - 0.219)*	0
Average		4.35 ± 1,05	0.511 ± 0.068	0.489 ± 0.061	0.045 ± 0.069	3.75 ± 3,86

[1]Nei, 1987; [2]Ibérico varieties.

III – Results and discussion

Information on pig native breeds is very scarce, even though they possess unique characteristics in terms of adaptation, hardiness, and quality of products, and could thus represent an interesting reservoir of genetic diversity (Cañón *et al.*, 2006; Foulley *et al.*, 2006; Peter *et al.*, 2007), in addition to being a major source of between-breed diversity . Knowledge of the structure of a livestock population in terms of sources of variability among and within breeds is essential for establishing conservation priorities and strategies (Caballero and Toro, 2002), with the long-term objective of maintaining genetic diversity for future generations (Notter, 1999). In the particular case of native breeds that have nearly become extinct, the possible existence of bottlenecks or admixture with other breeds in the recent past warrants special attention. Therefore, an assessment of their genetic diversity and possible relationships with other breeds represents a major step toward the development of conservation and improvement programs. Microsatellite markers are particularly suitable for genetic diversity studies, because of their large number, distribution throughout the genome, high level of polymorphism, codominant inheritance, neutrality with respect to selection, and easy automation of analytical procedures (Cañón *et al.*, 2001).

Although many of the breeds included in this study have been previously characterized genetically to some extent together with other breeds (Martínez *et al.*, 2000; Martínez *et al.*, 2003; Peinado *et al.*, 2003; Peinado *et al.*, 2006; Martínez *et al.*, 2007), a comprehensive analysis of the genetic diversity present in Spain and a source of important pig genetic resources has never been made. This study includes 6 of the 7 officially recognised local breeds and one population recently defined: Negro de Formentera e Ibiza. All the populations studied are endangered with the exception of two varieties of the Ibérico (Retinto and Entrepelado). Euskal Txerria and Celta breeds are representative of the Celtic trunk.

The total number of alleles found for the 24 microsatellite markers was 267, and polymorphisms in all loci were observed in most of the breeds. The number of alleles per locus ranged between 5 (S0215) and 25 (S0005), with a global mean for the 24 markers of 11.13 alleles per locus. This value is slightly superior to the range found in other European breeds of pigs (Laval *et al.*, 2000; San Cristobal *et al.*, 2006; Vicente *et al.*, 2008).

The within-breed analyses (Table 1) indicated that the mean number of alleles per locus was greatest in Retinto (6.38) and least in Euskaltxerria (3.00) with a mean number of alleles per locus over all populations of 4.35 ± 1.05. The average expected heterozygosity ranged between 0.596 in Celta and 0.369 for the Euskaltxerria breed. The proportion of loci that were not in HWE ($P < 0.05$) in each of the breeds analyzed ranged between 0 (Retinto Portugués, Mamellado, Celta) and 14 (Chato Murciano). Significant F_{IS} values were found in several populations, suggesting deviations from Hardy–Weinberg proportions might be due to consanguineous mating in those populations more endangered. Retinto, Torbiscal, Lampiño, Chato Murciano and Celta showed heterozygote deficit, while Retinto Portugués showed an excess of heterozygotes.

The F statistics showed a clear structure of the Spanish pig breeds. These statistics with their corresponding 95% confidence intervals obtained with 1000 bootstraps over loci were F_{IS} = 0.055 (0.030 – 0.085), F_{IT} = 0.254 (0.226 – 0.283) and F_{ST} = 0.211 (0.193 – 0.230). The large among-breed fixation index (F_{ST}) was significantly different from 0, indicating a clear structuring of the Spanish pig populations from the point of view of neutral genetic variability. This value is in agreement with the studies of Laval *et al*. (2000) and SanCristobal *et al.* (2006) on European breeds, but it markedly exceeds most previous studies of pig diversity (Martínez *et al.*, 2000; Yang *et al.*, 2003; Vicente *et al.*, 2008).

The graphical representation by Neighbor-joining of genetic distances between populations, calculated by the D_A distance, is shown in Fig. 1.

Low bootstrap values were obtained as it was expected when a high number of populations is analyzed (Felsenstein, 2004), and has been previously observed in the analysis of closely related domestic breeds with different genetic distances (Laval *et al.*, 2000; Martínez *et al.*, 2000; San Cristobal *et al.*, 2006; Vicente *et al.*, 2008). Nevertheless, all the Ibérico varieties formed a cluster while the rest of the breeds clustered separately. In the NJ tree, large branch lengths either corresponded to populations with low within population diversity. For example, the Euskaltxerria and the Negro Canario had very low observed and mean numbers of alleles and were found in the longest branches.

Bayesian clustering methods allow for the assignment of individuals to groups based on their genetic similarity and provide information on the number of ancestral populations underlying the observed genetic diversity. We tested values of K ranging from 2 to 16. The Ln Pr(X|K) increased sharply between K = 2 and K = 13, and stabilized between K=13 and K=16. These results would thus indicate that the appropriate value of K would be 13.

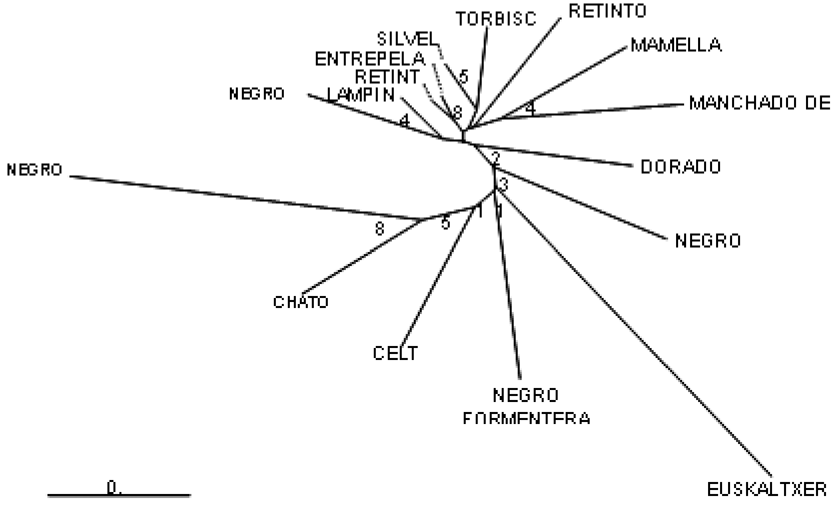

Fig. 1. Neighbour-joining dendrogram of 16 Spanish pig populations based on D_A (Nei 1983). Numbers are the percentage bootstrap values from 1000 replications of resampled loci.

The proportional membership of individual genotypes in the different clusters (Figure 2) indicates that the Iberian pig contains a high within-breed biodiversity as was previously reported by Martínez *et al*. (2000).

There are Ibérico varieties completely defined such as Retinto Portugués, Torbiscal, Negro de los Pedroches, Lampiño and Manchado de Jabugo. The rest of the Ibérico varieties belong to the "Red trunk" and it was difficult to separate them. Nevertheless, is was possible to discriminate between the Ibérico an the rest of the Spanish breeds. The Spanish breeds Iberian pig, Chato Murciano, Negro Canario, Negro de Formentera e Ibiza, Negro Mallorquín, Euskal Txerria and Celta showed that there was very little admixture between them, which are now quite distinct from each other. This result is in line with the F_{ST} estimate and confirms the distinctiveness of and low gene flow between the swine breeds analyzed, contrasting with results obtained when Structure was applied, for example, to horse (Vega-Pla *et al.*, 2006) and sheep (Quiroz *et al.*, 2008) populations, where admixture of breeds is very common.

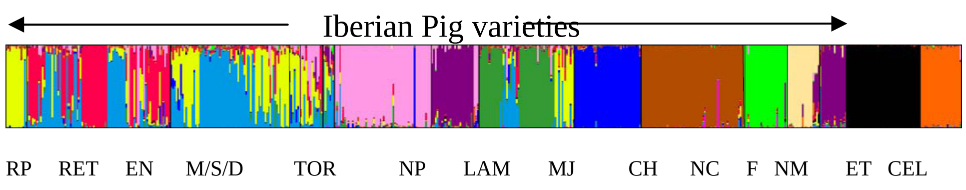

Fig. 2. Population structure of 16 Spanish pig populations using the model-based STRUCTURE software, where each animal is represented by a single vertical line divided into K colours, where K is the number of clusters assumed and the colours show the estimated individual proportions of cluster membership. Results are shown for K=13.

IV – Conclusions

There is a high porcine genetic diversity in Spain. The breeds sampled had a high level of differentiation, and most of them showed signs of accumulated inbreeding. Some of the breeds included in this study are involved in recent recovering processes and the results reported here may serve as useful indicators in setting conservation priorities, taking into consideration both among-population diversity and within- population variability, in addition to information on traits of current or potential economic importance, including adaptation.

Acknowledgements

The authors wish to acknowledge the financial support received by the Project RZ00-15 of the Programa Nacional de Recursos y Tecnologías Agroalimentarias of Ministerio de Ciencia y Tecnología of Spain.

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Effect of dietary oleic acid content: Different genetic regulation of fatty acid metabolism on muscle and fat of Iberian pigs

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Abstract. Feeding, genetics and their interactions influence animal tissue composition, thus affecting meat products quality. These influences are especially relevant in Iberian pig production. The goal of this work was the evaluation of the dietary oleic acid content effect on Iberian pigs muscle and fat composition and on δ9-desaturase (*SCD*) gene expression. Two groups of 13 castrated Iberian males were fed with two isocaloric diets differing on high-oleic sunflower oil content (0 *vs* 6%), from 28 to 110 kg live weight. Fatty acid profile of subcutaneous fat showed great differences between both groups. Nevertheless, intramuscular fat quantity and composition were not affected by the treatment. Results of qPCR show a great stability of *SCD* gene expression on adipose tissue. Contrarily, results obtained for muscle indicate a higher *SCD* gene expression (2x) on animals fed with 0% oleic diet (*P* <0.054), thus suggesting a different metabolic control of fat deposition on both studied tissues.

Keywords. Iberian pig – Oleic acid – *SCD* – Feeding.

Effets de la teneur en acide oléique de la ration : Différents contrôles géniques du métabolisme des acides gras du lard et de la longe chez les porcs Ibériques

Résumé. *L'alimentation, la génétique et leurs interactions influent sur la composition des tissus animaux et en conséquence sur la qualité de la viande. Cette influence est particulièrement importante chez un porc charcutier comme l'Ibérique. Dans ce travail, les effets de la teneur en acide oléique de la ration sur la composition lipidique et sur l'expression du gène SCD (δ9 désaturase) ont été étudiés dans la graisse de la longe et du lard des porcs Ibériques. Deux groupes de 13 mâles castrés furent nourris, de 28 jusqu'à 110 kg, avec des rations iso-caloriques avec différents pourcentages (0* vs *6%) d'huile de tournesol riche en acide oléique. Des différences significatives entre les deux groupes furent détectées dans le profil en acides gras du lard, mais la quantité et la composition de la graisse intramusculaire de la longe furent similaires. Les données obtenues pour l'analyse du transcriptome par qPCR montrent une grande stabilité de l'expression du gène SCD dans le tissu adipeux (lard). Les données obtenues dans le tissu musculaire (longe) indiquent cependant une plus grande expression (2x) de ce gène chez les porcs nourris avec l'aliment riche en acide oléique (P < 0,054). Le contrôle génique de la composition lipidique peut être différent entre les deux tissus.*

Mots-clés. *Porc Ibérique – Acide oléique – SCD – Alimentation.*

I – Introduction

Monounsaturated fatty acids (MUFA) enriched diets, through the inclusion of high oleic acid sunflower oil, are being used to fed Iberian-type pigs in order to mimic the profile of fatty acids (FA) characteristic of pigs fattened with the traditional system based on acorns from evergreen oaks and pasture. The effects of these enriched diets on the fatty acid composition of subcutaneous and intramuscular fat have been studied (Daza *et al*., 2005; Ventanas *et al*., 2008; Pérez-Palacios *et al*., 2009). In spite of the practical utilization of MUFA enriched diets in the production of Iberian pigs, their effect on the transcription of genes related to lipid metabolism has not been studied in this breed.

Stearoyl-CoA desaturase (*SCD*) gene has been proposed as a potential biomarker for fat deposition due to its key role in lipid metabolism and evidences of association between its expression and intramuscular fat content in pigs (Cánovas *et al.*, 2009; Zhao *et al.*, 2009). SCD is a key enzyme required for the biosynthesis of MUFA from saturated fatty acids (SFA) that are either synthesized de novo or derived from the diet. This enzyme catalizes the Δ^9-*cis* desaturation of a range of fatty acyl-CoA substrates, mainly palmitoyl and stearoyl-CoA, which are converted into palmitoleoyl- and oleoyl-CoA respectively. These products are the most abundant MUFA and serve as substrates for the synthesis of various kinds of lipids. Also, MUFA have been implicated as mediators in signal transduction and cellular differentiation. Thus, given the multiple roles of MUFA, variation in Stearoyl CoA desaturase expression or activity would be expected to affect a variety of key physiological variables in mammals. Also, in Iberian pig production, the influence of oleic acid content of meat products on the sensorial and technological quality is an added key variable that could be dependent on SCD function. For this reason this gene is considered a candidate to influence FA composition and pig meat and fat quality.

SCD is a highly regulated enzyme, which expression is known to be regulated by several dietary, hormonal and environmental factors (Paton and Ntambi, 2009). Recently, *SCD* expression has been shown to be down-regulated in response to linoleic and oleic acid treatment in vitro, in several species (Zulkifli *et al.*, 2010). This effect varies in intensity between tissues, cell types, species and fatty acids; and is a consequence of a reduction of *SCD* promoter activity in response to the fatty acid treatment. Specifically, the fatty acids are known to bind a highly conserved PUFA response region (PUFA-RR) of the *SCD* gene promoter, which contains several transcription factor binding sites as a sterol response element (SRE). Binding of FA to the PUFA-RR inhibits *SCD* transcription, with linoleic acid showing a more potent effect than oleic acid.

In live animals, changes in dietary fat composition have different impacts on the expression of genes related to lipid metabolism, and these impacts are highly dependent on the specie, age of the animal and studied tissue (Ding *et al*, 2003; Duran-Montge *et al*, 2009). *SCD* gene expression has been observed to be influenced by the FA dietary content (and specifically by dietary oleic acid) in adipose tissue and liver, but not in *semimembranosus* muscle of Duroc x Landrace female pigs of 100 Kg BW (Duran-Montge *et al*, 2009). Other tissue–specific responses of porcine *SCD* function have also been previously reported in dietary trials of protein reduction (Doran *et al*, 2006).

The objective of this work was to evaluate the effect of the oleic acid content of diet on fatty acid composition and transcription of *SCD* gene both in fat and muscle tissues of Iberian pigs. Also differences due to age, tissue and feeding status were studied.

II – Material and methods

1. Animals, dietary treatments and recorded traits

The experiment was performed at CIA *Dehesón del Encinar*. Twenty seven castrated males of the *Torbiscal* Iberian strain were used. At 28 kg of body weight the animals were housed individually, distributed in two groups fed with two different diets, one of them with the inclusion of high-oleic sunflower oil. Feeds composition is shown in Table 1. The energy content per kg of the feeds was: 3,130 Kcal crude energy for the control (C) diet, and 3,360 Kcal for the high oleic (HO) diet. Animals were fed twice a day. The higher energy content of the HO diet was compensated with a 10% increase in the feed quantity received by the animals of the control group. Water was provided *ad libitum*. Dietary treatment was maintained during 24 weeks until the pigs reached 110 kg of average live weight , when animals were slaughtered.

Live weight was recorded on live animals each two weeks during the treatment. Ham subcutaneous fat samples were obtained *in vivo* by shot-biopsies at different ages. One week before the sacrifice, fat biopsies were obtained immediately before and three hours after feeding. Animals were stunned and slaughtered at a local slaughterhouse (Alcaudete de la Jara, Toledo, Spain). At slaughter tissue samples were collected for gene expression analyses and stored at -80ºC. Backfat samples were taken at the level of the last rib and separated into outer and inner layers which were separately analyzed for fatty acid composition.

Table 1. Chemical and fatty acid composition of feeds

Diet	Control (C)	High oleic (HO)
Chemical composition, g/kg of feed		
Moisture	129.99	118.60
Lipids	15.33	74.98
Crude protein	127.00	140.00
Crude fiber	37.54	60.00
Nitrogen-free Extractives	505.56	399.38
Ash	41.60	48.10
Main Fatty acids, g/kg of feed		
C14:0	0.03	0.20
C16:0	2.19	5.85
C18:0	0.12	2.46
C18:1 n-9	1.52	44.78
C18:2	5.97	15.54

The extraction of total lipids from subcutaneous fat (inner and outer layers) and from *Longissimus dorsi*, and the analysis of fatty acid methyl esters by gas chromatography were performed according the procedures reported by Rey *et al*. (2006).

2. Gene expression analyses

RNA was extracted with Ribopure kit (Ambion), from 50-100 mg samples of different tissues from 13 animals of each experimental group: (i) ham subcutaneous fat biopsies obtained before and after feeding; (ii) inner layer of backfat samples from carcasses; and (iii) *Longissimus dorsi* muscle. RNA obtained was quantified with Nanodrop and evaluated with an Agilent Bioanalyzer. The quality of the obtained samples measured as 28S/18S ratio and RIN (RNA integrity number) was high.

SCD gene expression was quantified by qPCR with SYBR Green (Takara) in a Stratagene Real Time PCR (MxPro 3000). Two different primer pairs for quantification were designed using software QuickPri; in order to detect possible splice variants previously described for humans. The porcine mRNA sequence available was used for the primer design. These primer sets amplify fragments of 205bp and 218 bp and cover exons 4 to 5 and 5 to 6, respectively. Relative quantification was performed using *GADPH* as endogenous gene. All the reactions were run in triplets.

3. Statistical analyses

The influence of diet on fatty acid composition was separately analyzed for each fatty acid and tissue sample with a linear model fitting the mean, diet treatment and residual effects. A similar model was used for analysing *SCD* gene expression data fitting as covariable the expression values of the above quoted endogenous gene. All the analyses were performed using the GLM procedure of SAS 9.1 (SAS Institute Inc., Cary, NC, USA).

III – Results

1. Effects of diet on fatty acid composition

Both groups showed similar weights during all the experiment. The C group grew slightly faster than HO group, although this difference was not significant. At slaughter, mean carcass weight was 86.9 kg for the C group and 80.4 kg for the HO group.

To study the effect of diet on tissue composition, fatty acid profile of subcutaneous fat was analysed from samples obtained at slaughter (Table 2) and from fat biopsies obtained from live animals at 45 and 70 kg BW (results not shown). FA composition of subcutaneous fat showed significant differences between groups in all the samplings performed. Differences in FA composition of fat vary with sampling time and fat layer. Higher differences were obtained at slaughter on outer layer, with the HO diet group showing a greater percentage of MUFA (63.6 *vs* 54.8%; *P*<0.0001) and PUFA (9.2 *vs* 8.4%; *P*<0.007), and lower percentage of SFA (27.2 *vs* 36.7%; *P*<0.0001).

Table 2. Significant differences between Iberian pigs fed with Control (C) and High oleic (HO) diets for FA composition of outer and inner layers of subcutaneous fat samples obtained at slaughter

Backfat layer	Outer layer			Inner layer		
Fatty acid	**C diet**	**HO diet**	***P***	**C diet**	**HO diet**	***P***
C12:0	0.05±0.002	0.04±0.002	***	0.04±0.002	0.04±0.002	ns
C14:0	1.24±0.031	1.04±0.030	***	1.14±0.029	0.98±0.028	***
C16:0	23.85±0.224	18.83±0.216	***	25.11±0.225	20.49±0.217	***
C16:1	2.19±0.062	1.87±0.060	**	1.98±0.085	1.64±0.082	**
C17:0	0.40±0.019	0.31±0.018	**	0.38±0.020	0.26±0.019	***
C17:1	0.42±0.021	0.28±0.020	***	0.32±0.016	0.19±0.015	***
C18:0	10.84±0.228	6.67±0.219	***	13.53±0.384	9.15±0.370	***
C18:1	50.63±0.302	59.88±0.291	***	49.01±0.442	58.01±0.426	***
C18:2	6.88±0.149	7.65±0.144	**	5.72±0.117	6.48±0.113	***
C20:0	0.22±0.008	0.17±0.007	***	0.20±0.017	0.17±0.016	ns
C20:4	0.14±0.006	0.16±0.006	**	0.09±0.008	0.10±0.007	ns
C22:5	0.05±0.003	0.03±0.003	**	0.02±0.003	0.01±0.003	**
C22:6	0.02±0.001	0.03±0.001	**	0.01±0.003	0.01±0.003	ns

*** = *P*<0.001; ** = *P*<0.01; * = *P*<0.05; ns = not significant.

Fatty acids profile was also analysed in *Longissimus dorsi* muscle samples obtained at slaughter. In contrast to the backfat samples, the fatty acid profile of intramuscular fat was not affected by the dietary treatment (Table 3). Also total IMF content in this muscle was not different between both dietary groups.

Table 3. **Least-squares means and standard errors of FA percentages on intramuscular fat of Longissimus dorsi muscle samples from Iberian pigs fed with Control (C) and High oleic (HO) diets**

Fatty acid	C diet	HO diet	*P*	Fatty acid	C diet	HO diet	*P*
C10:0	0.10±0.017	0.09±0.016	ns	C18:3	0.22±0.009	0.22±0.009	ns
C12:0	0.07±0.002	0.08±0.002	ns	C18:4	0.05±0.002	0.05±0.002	ns
C14:0	1.26±0.032	1.28±0.031	ns	C20:0	0.16±0.007	0.16±0.007	ns
C16:0	24.03±0.297	24.23±0.286	ns	C20:1	0.72±0.018	0.72±0.017	ns
C16:1	3.93±0.242	3.78±0.234	ns	C20:3	0.16±0.009	0.16±0.009	ns
C17:0	0.25±0.014	0.25±0.013	ns	C20:4	1.70±0.137	1.54±0.132	ns
C17:1	0.22±0.012	0.22±0.012	ns	C20:5	0.21±0.018	0.20±0.017	ns
C18:0	10.57±0.288	10.95±0.277	ns	C22:5	0.15±0.016	0.15±0.015	ns
C18:1	50.40±0.606	50.33±0.584	ns	C22:6	0.07±0.006	0.05±0.006	*
C18:2	5.72±0.287	5.54±0.277	ns	% IMF	3.54±0.277	3.56±0.267	ns

** = P<0.01; * = P<0.05; ns = not significant

2. Effects of diet on gene expression: Fat and muscular tissues

The *SCD* gene expression was quantified in all backfat and loin samples taken from carcasses, and also in subcutaneous fat biopsies obtained before and three hours after feeding, one week before the slaughter of experimental pigs. Quantification was performed by using two different primer sets in the qPCR reactions, in order to account for possible expression differences specific of splice variants previously described for humans. The *SCD* quantity values obtained for both amplicons were highly correlated, indicating either the existence of only one transcript or more than one but with a highly correlated expression level. Thus, statistical analyses of the diet effect on *SCD* gene expression were performed using the expression values obtained with the primer set covering exons 4 to 5, which potentially include all the possible transcript variants.

The *SCD* gene expression in fat tissues was not affected by the dietary fat neither by the feeding status of the animal (Figure 1). The expression level of this gene was higher in the biopsies obtained one week before slaughter, than in the sampling performed from the carcasses. This could be probably due to the effect of the sampling region or the fat layer used for the RNA extraction, backfat inner layer at slaughter, and mainly ham outer layer on the biopsies.

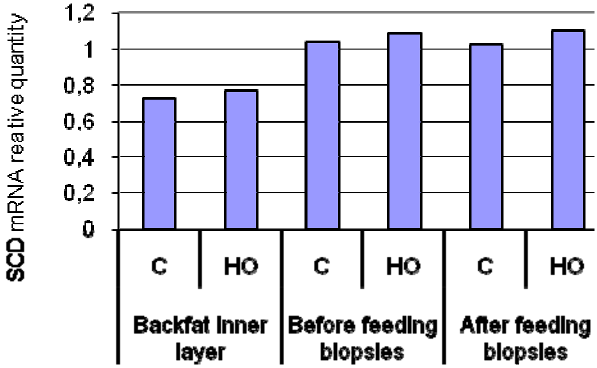

Fig. 1. **Mean SCD gene expression in subcutaneous fat samples from different samplings of control (C) and high oleic (HO) groups.**

SCD gene expression in muscle was significantly affected by the dietary treatment (Figure 2). A higher expression (2x) was observed on animals fed with control diet. The mean expression values were 0.249 ± 0.040 and 0.126 ± 0.039 for C and HO groups, respectively (P <0.054).

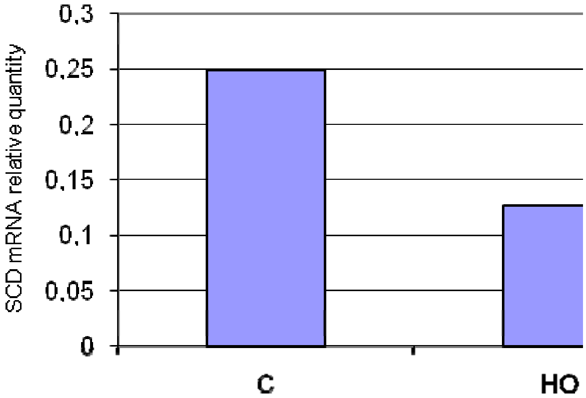

Fig. 2. Mean SCD gene expression in Longissimus dorsi samples of pigs from control (C) and high oleic (HO) diet treatments.

IV – Discussion

Feeding, genetics and their interactions influence animal tissue composition, thus affecting meat products quality. These influences are especially relevant in Iberian pig production, in which diet composition and genetic type are the strongest determinants in their meat and fat high quality and ability for processing. In fact, the highest quality and prize of Iberian pig meat products are obtained from pure animals reared outdoors, implying the consumption of acorns and grass (López-Bote, 1998). Due to the restricted acorn production and the high demand of Iberian pig products, the use of MUFA enriched concentrates is becoming a usual practice in order to achieve FA profiles in animal tissues similar to that obtained with the consumption of acorns, as the fatty acid profiles of animals tissues are expected to reflect the fatty acid composition of the feeds received (Daza *et al*., 2005; Ventanas *et al*., 2008; Pérez-Palacios *et al*., 2009).

Nevertheless, the effects of feeding with MUFA enriched diets on the different animal tissues composition are not clear. Previous studies show non consistent and tissue-specific effects although their comparison is difficult due to the different dietary treatments (high or moderate inclusion of MUFA in enriched diet) or tissue (muscle type) and sampling. Moreover, the relative roles of direct deposition of dietary FA and endogenous synthesis and the regulation of FA synthesis by diet components are key factors in pig lipid metabolism and production, which are not well understood.

In the present work, we studied the effect of a long-term moderate MUFA inclusion in the diet on loin muscle and subcutaneous fat FA composition. Animal tissue analyses showed no effect of dietary fat on muscle fatty acid composition. Both C and HO groups showed similar intramuscular FA pattern instead of the very different FA composition of their diets. In contrast, subcutaneous fat FA composition clearly reflected the dietary FA composition, with much higher MUFA and lower SFA content in the HO group. Slightly higher PUFA content was observed in the HO group which may be attributed to the positive effect of dietary fat on C18:2 content. This increase of C18:2 percentage has been reported as a potentially undesirable secondary effect of MUFA enriched diets by Ventanas *et al*. (2008), although their MUFA inclusion level and the corresponding C18:2 increase were much higher than those of the present study.

The different response of adipose and muscular tissues to the dietary influence has been previously observed with a similar pattern in other works, that is, adipose tissue reflect to a greater extent the dietary modifications in terms of total tissue FA content, but the influence of dietary treatment on muscle FA is not so evident (Duran-Montge *et al.*, 2009). In Iberian pigs, Pérez-Palacios *et al*. (2009) reported significant effects on backfat and *Semimembranosus* muscle but not on *B. femoris*.

Regarding the effect of the diet on *SCD* gene expression, no difference on mRNA abundance was observed between fat samples of C and HO animals. Previous results of differential gene expression obtained from this experimental material with a microarray approach indicated that changes in backfat gene expression induced by fatty acid composition of the diet are small in number and magnitude (Óvilo *et al.*, 2009). This result indicate a low genetic responsiveness of fat tissues to dietary treatments which could be particularly relevant in obese genetic types, as a consequence of a dilution effect, as it has been described in humans (Van Erk *et al.*, 2008).

In contrast to fat tissue, muscles show a much different response to the dietary treatment not only at the tissue composition but also at *SCD* transcriptional levels. Loin samples are not affected in this study by the dietary fat, showing identical FA and total IMF composition between groups. This fact would reflect an effect at the transcriptional or enzymatic activity levels that lead to an increase in MUFA endogenous synthesis or desaturation thus compensating the direct deposition of dietary FA. The results of *SCD* gene expression quantification agree with this hypothesis, as the C animals show twice *SCD* gene expression than HO animals. Although there are abundant evidences of tissue-specific responses of lipogenic enzymes expression in response to dietary treatments, the reasons of these changes are not clear. One possible explanation could be the different levels and/or classes of transcription factors regulating the enzyme expression, which accompany the morphological and metabolic differences between both intramuscular and subcutaneous fat depots (Gardan *et al.*, 2006; Gondret *et al.*, 2008).

Our work support previous evidences indicating the modulation of *SCD* gene expression by dietary fatty acids and also the tissue-specificity of this effect. Nevertheless, the higher influence would be expected to appear in the most lipogenic tissues, which in pigs are liver and adipose tissue (Duran-Montge *et al.*, 2009). The higher transcriptional response obtained in muscle in our work could be related to the higher lipogenic potential of Iberian muscle tissue in comparison with other breeds. The study of these dietary effects in other genetic backgrounds is indicated in order to understand the effects detected.

Acknowledgments

The research was supported by INIA grant RTA2007-00075-00-00.

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Δ9-desaturase polymorphism association with fatty acid profile of Italian PDO dry cured hams

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Abstract. PDO dry-cured ham (prosciutto) is the main product of the Italian pig industry. Fatty acid profile is of major importance for the technological properties of the fresh thigh and for the dietary and sensory quality of the ham. Δ9-desaturase (SCD) is a microsomal enzyme required for the biosynthesis of unsaturated fatty acids and therefore a potential candidate gene to explain the differences in the MUFA content in meat and fat. Through sequencing, a SNP was observed in the SCD promoter. Forty-six pigs of Italian Landrace x Italian Large White cross breed ("reference hybrids") and 32 Goland, a commercial hybrid, were genotyped by PCR-RFLP for this mutation. Fatty acid profiles were determined by gas chromatography in thigh subcutaneous fat. Single locus genotype association analysis was carried out using the General Linear Model in SPSS. The genotypic frequencies of the SCD polymorphism were different between the two pig hybrids. However, independently of the pig genetic type, significant associations (P < 0.05) were found between the SCD polymorphism and the ratios of palmitoleic to palmitic acids and of oleic to stearic acids, as well as the ratio of MUFA to SFA. These findings confirm our previous results and suggest that the SCD promoter polymorphism is associated with the quality of PDO dry cured Italian hams.

Keywords. Italian heavy pig – SCD – Fatty acid profile – Association – Dry-cured ham.

Association entre un polymorphisme de la Δ9-désaturase et le profil en acides gras des jambons crus AOC italiens

Résumé. *Le principal produit de l'industrie porcine italienne est le jambon cru AOC. Le profil en acides gras est d'importance majeure pour les propriétés technologiques de la cuisse et pour la qualité diététique et sensorielle du jambon. La Δ9-désaturase (SCD) est une enzyme microsomale impliquée dans la biosynthèse des acides gras insaturés ce qui en fait un gène candidat potentiel pour interpréter les différences dans le contenu en MUFA de la viande et de la graisse. Un SNP a été identifié par séquençage dans le promoteur de la SCD. Quarante-six porcs Landrace Italien x Large White Italien (hybrides de référence) ainsi que 32 porcs Goland, un hybride commercial, ont été génotypés pour cette mutation par PCR-RFLP. La composition en acides gras dans la graisse sous-cutanée de la cuisse a été déterminée par chromatographie gazeuse. L'analyse d'association entre le génotype et le profil en acides gras a été réalisée par le General Linear Model de SPSS. La fréquence génotypique pour cette mutation de la SCD était différente entre les deux groupes de porc étudiés. De plus, indépendamment de la souche porcine, des associations significatives ont été observées (P<0,05) entre le polymorphisme du promoteur de la SCD et les ratios entre acides palmitoléique et palmitique, entre acides oléique et stéarique, ainsi qu'entre MUFA et SFA. Ces résultats confirment nos observations antérieures et suggèrent que le polymorphisme du promoteur de la SCD est associé à la qualité du jambon cru italien AOC.*

Mots-clès. *Porc lourd italien – SCD – Profil en acides gras – Association – Jambon cru.*

I – Introduction

PDO dry-cured ham (prosciutto) is the main product of the Italian pig industry and more than

80% of the pig production is destined to the PDO traditional Italian ham market (Renaville *et al.*, 2010). To obtain high quality PDO dry-cured hams, the production is subjected to rules fixed by several Consortia (Bosi and Russo, 2004) concerning the characteristics of fresh thighs, genetic type, feedstuff, age and slaughtering weight of animals. Hams are obtained from pigs of at least 9 months of age and 160 kg of minimum live weight. Heavy pigs have two main origins: the first is a specific Italian selection obtained from the "traditional breeds" genetically improved by the Italian Breeders Association, in particular the crossing breeds used are Italian Large White, Italian Landrance, Italian Duroc and their crosses are called "reference crosses". The second one regards commercial hybrids produced by international breeding companies in agreement with the objectives of the Italian heavy pig selection.

It is widely accepted that the genetic type can influence ham quality (Vitale *et al.,* 2009) and fatty acids profile (Lo Fiego *et al.*, 2005; Wood *et al.*, 2008; Piasentier *et al.*, 2009). In particular unsaturated fatty acids are more susceptible to oxidation than saturated fatty acids and they can influence the sensory quality and acceptability of dry cured hams (Musella *et al.,* 2009).

Δ9 desaturase, also called Stearoyl-CoA desaturase (SCD), is a rate-limiting enzyme located in the membrane of the endoplasmic reticulum and it is responsible for the insertion of a double bond between carbons 9 and 10 into saturated fatty acids (Ntambi and Miyazaki, 2004). Previously Ramos *et al.* (2008), considering another SNP, showed that a SCD polymorphism was associated with colour, weight and yield of cured hams.

The purpose of this work was to estimate the effect of the SCD polymorphisms on fatty acid composition of subcutaneous adipose tissue of thighs destined to the production of PDO Italian dry-cured hams.

II – Materials and methods

Forty six thighs of Italian Landrace x Italian Large White cross pigs ("reference hybrids", IL x LW) and 32 thighs of Goland pigs (commercial hybrid) were considered. Pigs were reared in the same farm and fed with a standard cereals-soybean based meal as described in Minelli *et al.* (2010). All the thighs were destined to the production of PDO Italian dry-cured hams.

Fatty acid profile of subcutaneous fat was determined using a TRACE™ GC Ultra (Thermo Electron Corporation, Rodano, Milano, Italy) equipped with the Ultra Fast Module (UFM), a Fast Flame Ionization Detector and a UFM-Carbovax column, 5 m long, 0.1 mm i.d, 0.2 μm film thickness as described in a previous paper (Ficarra *et al*., 2010). The FAME were identified by comparison of each retention time with the known retention times of the corresponding pure standards (Supelco 37 Component FAME mix and PUFA standard n. 2, animal source, Supelco, Bellafonte, PA, USA). For quantification purposes, the response factor was calculated and the method of the internal standard was used. Results were expressed as g of each fatty acid methyl ester/100 g of total lipids.

DNA was extracted from muscle samples using a standard DNA extraction method. The genotype procedure was as follows. The PCR amplifications were performed using 12.5 ng of porcine DNA, 1 x PCR buffer, 0.125 mM each dNTP, 0.3 mM of each primer and 0.35 U Taq polymerase (Promega, Madison, WI, USA). The primers used were: forward: CTCTGTCTCCTCCCCTCTCC; reverse: GATCACTTTCCCAGGGATGA. PCR products were sequenced using an ABI automated DNA sequencer (Applied Biosystems, Foster City, CA, USA). A C/T polymorphism was found in the fragment and a PCR-RFLP test was designed using the specific restriction enzyme PflF1 following the recommendations of the manufacturer. The fragment size was 322 bp for allele 1 and 212 + 110 bp for allele 2. With the aim of estimating the SCD enzyme activity Δ9 desaturase index was assessed and calculated as proposed by Smith *et al.* (2002), (16:1 + 18:1) / (14:0 + 16:0 + 18:0 + 16:1 + 18:1).

The statistical analyses were done using the package SPSS vers. 17 (SPSS Inc., Illinois). The difference in SCD genotypic frequencies between hybrids was tested by chi-square tests. Fatty acids profiles were subjected to analysis of variance using a two way factorial design with hybrids and SCD genotype as fixed effects. Interaction between hybrids and SCD genotype was considered, but the value was not reported in Table 2 because it never reached the level of significance ($P > 0.05$). Post hoc pairwise comparisons were analyzed by the Tukey-Kramer tests.

III – Results and discussion

The SCD promoter genotypes were different between hybrids (Table 1). In particular Goland commercial hybrid pigs had a higher frequency of genotype 22 than IL x LW pigs (84.3% *vs*. 50.0%). Conversely the IL x LW pigs presented a higher frequency of genotype 12 than Goland pig (37.0% *vs*. 15.6%) in which the genotype 11 were not found. Renaville *et al.* (2010) reported similar frequencies (89% for 22 and 11% for 12) in a Goland population (n = 149). Overall, these authors reported a frequency of genotype 22, 12 and 11 of 76, 22 and 2% respectively, considering 615 pigs from four different commercial hybrids destined to the PDO traditional Italian ham market. Ramos *et al.* (2008) in a study that took into account 321 US country hams derived from commercially crossbred pigs found a frequency of SCD markers genotype 22, 12 and 11 of 50, 39 and 11% respectively. Although, the polymorphism of these 2 studies was different, it is in completely linked with our mutation in the promoter allowing to compare the results.

Table 1. Genotypic frequencies of Stearoyl Co-A desaturase (SCD) markers in different pig hybrid - Italian Landrance x Italian Large White (IL x LW) and Goland.

Marker	Hybrids		Total	*P*-value
	IL x LW	Goland		
No of thighs	46	32	78	
SCD marker				0.01
11	6	0	7.7 %	
12	17	5	28.2 %	
22	23	27	64.1 %	

The estimated marginal means, i.e. the mean response for each factor adjusted for the other variables in the model, of the fatty acid composition of the subcutaneous adipose tissue of thighs are reported in Table 2. A significant effect of genetic type was observed. The IL x LW "reference hybrid" had significantly higher proportion of C18:0 ($P < 0.01$), C20:0 ($P < 0.01$), C18:1 ($P < 0.01$), C20:1 ($P < 0.01$) and had, consequently, higher levels of saturated fatty acids (SFA) ($P < 0.01$) and monounsaturated fatty acids (MUFA) ($P < 0.01$) than Goland commercial pigs. The MUFA / SFA ratio was not affected by the hybrid ($P > 0.05$). Goland commercial hybrids showed significantly higher proportion of C18:2 ($P < 0.01$), C18:3 ($P < 0.01$), C20:2 ($P < 0.01$), and had, consequently, higher level of polyunsaturated fatty acids (PUFA) ($P < 0.01$) than "reference hybrids". In particular Goland hybrids had a content of linoleic acid (C18:2), 12.38%, near to the threshold value set by the Consortia for this fatty acid (15%) and higher than those proposed by Girard *et al.* (1988) (12%). This value was introduced by the Consortia with the aim to limit the content of PUFA of the subcutaneous adipose tissue of fresh thighs in order to guarantee an adequate firmness of fat and to limit its oxidability (Bosi and Russo, 2004). Our results are consistent with the results obtained by Lo Fiego *et al.* (2005) that considered 112 pig, 56 "traditional pigs" (Italian Landrace x Large White cross) and 56 animals from a commercial hybrid, destined to the production of PDO traditional dry-cured ham. The above cited authors suggested that the genetic selection of the commercial hybrids have significantly increased pigs' performance reducing the quantity of carcass fat with an increasing in PUFA content. Consequently, it makes the thighs less suitable for curing.

Table 2. Effect of the different pig hybrid –Italian Landrance x Italian Large White cross (IL x LW) and Goland– and of Stearoyl Co-A desaturase (SCD) markers on thickness (mm) and on fatty acids composition (g/100g of total lipids) of subcutaneous adipose tissue of thighs (estimated marginal means).

	Hybrids		SCD markers			RMSE	P – value	
	IL x LW	Goland	11	12	22		H	SG
Fat thickness	25.41	23.95	27.00	25.71	22.85	5.513	0.713	0.171
C14:0	1.21	1.22	1.17	1.25	1.20	0.146	0.759	0.366
C16:0	21.90	21.12	21.76	21.74	21.34	1.447	0.072	0.666
C16:1	2.01	2.28	1.82	2.17	2.22	0.294	0.075	0.078
C17:0	0.43	0.38	0.39	0.40	0.42	0.116	0.078	0.522
C17:1	0.41	0.38	0.36	0.39	0.43	0.113	0.163	0.172
C18:0	13.49^{A}	10.99^{B}	14.21^{a}	12.27^{b}	11.84^{b}	0.933	0.000	0.014
C18:1	41.72^{A}	38.05^{B}	41.03	39.44	40.67	2.285	0.000	0.148
C18:2	8.56^{B}	12.38^{A}	7.95	10.44	10.80	1.490	0.000	0.287
C18:3n-6	0.13	0.12	0.12^{AB}	0.10^{B}	0.15^{A}	0.054	0.345	0.005
C18:3	0.40^{B}	0.55^{A}	0.37	0.46	0.50	0.077	0.000	0.084
C20:0	0.20^{A}	0.16^{B}	0.21	0.17	0.18	0.028	0.001	0.080
C20:1	0.94^{A}	0.73^{B}	1.04^{A}	0.75^{C}	0.87^{B}	0.143	0.001	0.005
C20:2	0.44^{B}	0.54^{A}	0.45	0.46	0.52	0.076	0.000	0.063
C20:3	0.08	0.08	0.08	0.08	0.09	0.022	0.476	0.273
C20:4	0.13	0.15	0.12	0.14	0.15	0.032	0.057	0.429
SFA†	37.22^{A}	33.86^{B}	37.74	35.83	34.99	2.151	0.000	0.337
MUFA††	45.08^{A}	41.44^{B}	44.25	42.75	44.19	2.424	0.000	0.095
PUFA†††	9.73^{B}	13.82^{A}	9.08	11.67	12.21	1.622	0.000	0.229
C16:1 / C16:0	0.09^{B}	0.11^{A}	0.08^{b}	0.10^{a}	0.10^{a}	0.011	0.001	0.025
C17:1 / C17:0	0.96	1.03	0.91	0.99	1.03	0.107	0.217	0.156
C18:1 / C18:0	3.11^{b}	3.51^{a}	2.90^{b}	3.27^{a}	3.46^{a}	0.336	0.010	0.021
C20:1 / C20:0	4.80	4.72	4.89	4.62	4.87	0.714	0.858	0.522
Δ9 desaturase index††††	0.545	0.548	0.536^{b}	0.542^{ab}	0.556^{a}	0.018	0.813	0.025
MUFA / SFA	1.21	1.23	1.17^{b}	1.20^{b}	1.27^{a}	0.097	0.878	0.035

RMSE: root mean standard error; H: effect of hybrid; SG: effect of SCD markers; ^{A,B,C} Means within the same row with unlike letters differ significantly at $P \leq 0.01$; ^{a,b,c} Means within the same row with unlike letters differ significantly at $P \leq 0.05$.
†SFA (saturated fatty acids) = C14:0 + C16:0 + C17:0 + C18:0 + C20:0.
††MUFA (monounsaturated fatty acids) = C16:1 + C17:1 + C18:1 + C20:1.
†††PUFA (polyunsaturated fatty acids) = C18:2 + C18:3n-6 + C18:3 + C20:2 + C20:3 + C20:4.
††††Δ9 desaturase index = (C16:1 + C18:1) / (C14:0 + C16:0 + C18:0 + C16:1 + C18:1)

The "reference hybrids" obtained from the "traditional breeds" had lower C18:1 / C18:0 ($P < 0.05$) and C16:1 / C16:0 ratio ($P < 0.01$) than Goland hybrid, however the Δ9 desaturase index was similar between breeds ($P > 0.05$).

Stearoyl-CoA Desaturase or Δ9-desaturase is a key enzyme in lipid metabolism, indeed, it is involved in the conversion of SFA in MUFA. In particular SCD enzyme converts C16:0 and C18:0 to C16:1 and C18:1 respectively. Moreover Cánovas *et al.* (2009) suggested that SCD might be an effective potential biomarker for fat deposition in pigs. Renaville *et al.* (2010) found an association between SCD polymorphism and ham fat thickness and the lowest value was recorded by the genotype 11.

In our trial the effect of the SCD promoter genotype on ham fat thickness did not reach the threshold of significance ($P > 0.05$) likely due to the little number of pigs considered. Significant associations were found between the SCD markers and the levels of C18:3n-6 and C20:1. Genotype 12 showed the lowest level of C20:1 ($P < 0.01$) which is probably synthesized by elongation of oleic acid (Durant-Montgé *et al.,* 2010). Genotype 22 presented significantly the highest C18:3n-6 level ($P < 0.01$) although the SCD enzyme is not directly involved in the metabolism of C18:3n-6, the effect observed might be due to an indirect effect of SCD. Indeed, SCD is involved in the packaging of fatty acids in the Very Low Density Lipoprotein (Stefan *et al.*, 2008) and therefore in their transport to the adipose tissue. An increase activity of SCD might increase the trafficking of C18:3. The content of linoleic acid (C18:2) in thigh subcutaneous fat was not affected by SCD marker genotype, and it is lower than the value fixed in the rules set by the Consortia (15%).

Moreover genotype 11 presented the highest C18:0 ($P < 0.05$) content and the lowest C16:1 / C16:0 ($P < 0.05$) and C18:1 / C18:0 ratio ($P < 0.05$). In general, these results highlighted a lower SCD enzyme activity of genotype 11. This statement seems to be confirmed by the Δ9 desaturase index that was lower in genotype 11 than genotype 22 ($P < 0.05$). This index was previously used as an estimator of SCD enzyme activity in cattle and in pig (Corl *et al.,* 2001; Smith *et al.,* 2002). The total content of SFA, MUFA and PUFA were not affected by SCD genotype, however the genotype 22 presented higher MUFA / SFA ratio than the other groups ($P < 0.05$) probably in consequence of its higher SCD enzyme activity. This result suggests that the lipids in the ham of pig with a SCD promoter genotype 22 have a higher nutritional quality (WHO, 2003) than those of the other genotypes.

IV – Conclusions

In summary we can be concluded that there are differences in the genotypic frequencies of the SCD promoter polymorphism between the two pig hybrids. In particular, the “reference hybrids” have lower frequencies of genotype 22 and higher frequencies of genotypes 11 and 12 than Goland commercial hybrids. However, independently of the pig genetic type, SCD promoter genotype 11 had the lowest ratios of palmitoleic to palmitic acid and of oleic to stearic acid. Accordingly the genotype 22 had the highest ratio of MUFA to SFA and Δ9 desaturase index. These findings suggest, in agreement with our previous results, that the SCD promoter polymorphism is associated with the quality of PDO Italian dry cured hams probably reflecting a different SCD enzyme activity.

Acknowledgements

The authors would like to thank Dr. Elena Saccà for her technical assistance. The research was supported by MIUR- PRIN 2007 (Prof. E. Piasentier).

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New genetic tests to select Iberian pigs

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Abstract. One of the main tasks in animal breeding is the identification of genes controlling economically important traits. Several polymorphisms (SNPs) in genes associated with growth, fatness, meat quality, disease resistance and prolificacy have been described in pigs and some of these SNPs have been incorporated in breeding programmes of pig industry. However, most of these SNPs are not functional and therefore they may not show any phenotypic effect in a particular population. Besides, most of them are fixed or at low frequency in *Iberian* pigs. We propose a new approach to identify new SNPs in complex candidate genes for designing useful genetic tests to select *Iberian* pigs. As an example, we have selected, from the PorcineSNP60 BeadChip genotype information, one panel of 96 SNPs flanking *FAT1* SSC4 QTL, Leptin Receptor (*LEPR*) and Acetyl Coenzime-A Carboxilase (*ACACA*) genes regions. A total of 334 *Iberian* pigs from AECERIBER performance tests were genotyped with this panel. Significant effects of five SNPs on growth, ham and loin weight were detected. The ability of this approach to identify genetic markers useful as a tool for selecting *Iberian* pigs is discussed.

Keywords. Iberian pig – Genetic tests – Selection.

Nouveaux tests génétiques pour la sélection des porcs Ibériques

Résumé. *Une des principales activités en amélioration génétique animale est l'identification de gènes responsables de caractères d'intérêt économique. Plusieurs polymorphismes (SNPs) localisés dans des gènes associés avec la croissance, l'engraissement, la qualité de la viande et la prolificité ont été décrits chez le porc. Certains de ces SNPs, conjointement avec les informations de performances, sont utilisés par la filière porcine. Toutefois, la plupart de ces SNPs ne sont pas fonctionnels, et selon la population étudiée, n'ont pas systématiquement un effet phénotypique. De plus, la plupart d'entre eux sont fixés ou ont une fréquence faible chez les porcs Ibériques. Nous proposons une nouvelle approche pour identifier de nouveaux SNPs dans des gènes candidats complexes, et pour réaliser des tests génétiques utiles à la sélection des porcs Ibériques. Nous avons sélectionné, à partir des résultats de génotypage de la puce PorcineSNP60, un panel de 96 SNPs flanquant le QTL FAT1 et les régions des gènes du Récepteur à la Leptine (LEPR) et de l'Acétyl-CoA Carboxylase (ACACA). 334 porcs Ibériques issus du programme de tests de performances AECERIBER ont été génotypés grâce à ce panel. Des effets significatifs de cinq SNPs ont été détectés pour les caractères de croissance, et de poids de jambon et de filet. La capacité de cette approche à identifier des marqueurs génétiques utiles à la sélection des porcs Ibériques est discutée.*

Mots-clés. *Porc Ibérique – Tests génétiques – Sélection.*

I – Introduction

In the last decades, one of the main tasks in animal breeding has been to identify genes controlling production traits. Single nucleotide polymorphisms (SNPs) have become the genetic markers of choice because they are the most frequent type of sequence variation of DNA being distributed over all the genome. In pigs, several polymorphisms in genes associated with diverse economic and morphological traits have been described and, some of them, together with traditional performance information, have been incorporated in breeding programmes of the pig industry. However, most of these SNPs were detected from quantitative trait loci (QTL) experiments in crosses between divergent breeds of pigs (Hu *et al*., 2007) and their effects need to be validated in commercial populations since they may not be the causal mutations of the phenotypic changes. Besides, the most relevant associations between SNPs and traits reported in porcine breeds have not been confirmed in *Iberian* pigs. In some cases this is due to the fact that the studies are based on SNP present at low frequency or monomorphic in this breed (Pérez-Montarelo *et al*., 2010).

Two experimental crosses involving *Landrace* or *Meishan* as the maternal breed and Guadyerbas *Iberian* strain as the paternal line have been analyzed during the last years in order to identify chromosomal regions affecting economically important traits, particularly those related to growth, carcass composition, meat quality and reproduction (Varona *et al*., 2002; Noguera *et al*., 2009). These studies allowed detecting QTL located on chromosomes 4, 6 and 12 with important effects on live weight, back fat thickness and meat and fat quality traits (Pérez-Enciso *et al*., 2000; Ovilo *et al*., 2000; Muñoz *et al*., 2007).

New molecular technologies, such as the emerging high-throughput genotyping have allowed the development of high-density SNP genotyping platforms which are useful to identify mapped allelic variants in livestock animals. We propose to take advantage of these technologies for identifying genetic markers useful as a complementary selection tool for *Iberian* pigs. The first objective of this work was to build up a low-density panel of polymorphic SNPs mapped in the three quoted chromosomal regions, using the PorcineSNP60 BeadChip (Ramos *et al*., 2009. The second objective was to identify SNP effects on production traits in a representative sample of the *Iberian* pig breed, using the low-density panel developed.

II – Materials and methods

1. Animals and phenotypic records

Since 1993, the Spanish Association of *Iberian* Pig Breeders (AECERIBER) has carried out annual tests of carcass performance based on family groups of animals sampled at weaning from different herds which are transferred to a common testing farm. The pigs were under restricted feeding with concentrates until they reached a live weight of approximately 100 kg. Afterwards, they were fattened in 'Montanera' until an average weight of 160 kg. The live weight at the day before the slaughter and the weight of the warm carcass, trimmed hams, forelegs and loins, were individually registered. Samples of *longissimus dorsi* muscle at level of the fourth rib were collected to determinate, using near infrared spectroscopy (NIRS), the content of intramuscular fat, moisture and protein (Fernández *et al*., 2003).

Available records came from 334 barrows born in eight breeding nucleus from South-western Spain that maintain pigs representing the main morphological types of the breed. Data were collected from 2002 to 2005 and were grouped in four different batches according to the slaughter date. The elementary statistics of the analysed traits are reported in Table 1. Sire and dam identification was available for all animals included in the study.

2. SNP genotyping

Genomic DNA was extracted from blood or meat samples using a standard protocol. In a first step, samples of 26 *Iberian* pigs from 16 breeding nuclei registered in the Herd Book were genotyped for 62,163 SNPs using the Porcine SNP60 BeadChip (Illumina, San Diego, CA, USA). Genotyping reactions were performed on an "Infinium DNA Analysis Assay" at the Veterinary Service of Molecular Genetics (Universitàt Autonoma de Barcelona, Spain). We selected 85 SNP probes, from the above genotype information, with minor allele frequencies greater than 0.20 and overlapping the targeted regions: *FAT1* on chromosome 4 (SSC4), Leptin Receptor (*LEPR*) on SSC6 and Acetyl Coenzime-A Carboxilase (*ACACA*) on SSC12. These regions were selected because *FAT1* region corresponds to the main QTL region on SSC4 where several genes of the *FABP* family, related to lipid metabolism in different tissues are located (Szczerbal *et al*., 2007), *LEPR* gene has been studied as a candidate to underlay the main QTL on SSC6 for growth and body composition traits (Ovilo *et al*., 2005; Morhman *et al*., 2006) and *ACACA* is a key gene in fatty acid metabolism and it maps in the central region of the

QTL on SSC12 for fatty acid content of porcine back fat (Gallardo *et al*., 2007; Muñoz *et al*., 2007).

Table 1. Main statistics for the analysed traits

Trait	N	Mean	SD	Minimum	Maximum
Growth in extensive fattening period					
Starting weight, kg	171	98.38	10.58	73.00	121.00
Final weight, kg	325	162.88	13.39	131.00	225.00
Carcass weight, kg	325	129.33	11.08	106.50	178.50
Premium cuts yield					
Hams, kg	327	21.39	2.01	16.90	30.80
Forelegs, kg	329	14.52	1.37	11.50	19.70
Loins, kg	333	2.95	0.40	2.00	4.48
Meat quality					
Intramuscular fat, %	325	9.74	2.93	2.54	22.10
Moisture, %	324	69.39	2.28	56.46	74.44
Protein content, %	325	19.44	1.16	17.08	22.70

We increased the above set of 85 SNPs with eleven additional SNPs probes, which correspond to *LEPR* SNPs (five probes) and *ACACA* SNPs (six probes), previously identified by sequencing. The 96 SNPs covered a region of 146,146,887bp: 13,332,046bp on *FAT1* region of SSC4 (between INRA0014028 and ALGA0025271 probes), 13,020,811bp on SSC6 *LEPR* region (between ASGA0029534 and ALGA0037285) and 2,606,730bp on SSC12 *ACACA* region (between ALGA0066258 and ALGA0108055). The 96 finally selected SNPs loci were simultaneously interrogated with the GoldenGate Genotyping Assay.

3. Statistical analysis

Individual SNP effects on phenotypes were estimated fitting animal models, which besides the random polygenic effects included the following fixed effects: batch, additive and dominant SNP effects and the carcass weight as a covariate for premium cuts yield and meat composition traits. The analyses were performed using the Qxpak v.4 software (Pérez-Enciso and Misztal, 2004). The statistical significance of each SNP effects was tested comparing the full and reduced models by means of $\chi 2$ approaches to the distribution of log-likelihood ratios (LR). This quantity is distributed as a $\chi 2$ with degrees of freedom (d.f.) equal to the difference in the number of parameters between the alternative and the null models. We used one additional d.f. to calculate P-values stricter than the nominal ones. This extra d.f. accounts for the position of multiple SNPs tested.

III – Results and discussion

Seven out of the 96 selected probes failed to produce amplification in all samples: one out of the 30 located on SSC4, four out of the 35 corresponding to SSC6 and two out of the ones mapped on SSC12. Moreover, four of the five *LEPR* probes and one of the six *ACACA* probes were monomorphic. Sixty of the 89 probes showed SNPs allelic frequencies between 0.30 and 0.70. The average genetic distances between the genotyped SNPs was approximately 0.34 cM (340 kb, Build9) and the average value of linkage disequilibrium (LD) between adjacent SNPs was r 2 = 0.10.

SNP effects on several growth and carcass traits have been found in the three chromosomal regions analysed (Table 2). The most remarkable results corresponded to ALGA0123491 SNP probe mapping on SSC12. This SNP was associated with the live weights at the start and end of the extensive fattening period and also with the carcass weight. Additive effects explained about 2% of the mean of these traits. Dominant effects of similar magnitude were detected for final live weight and carcass weight. A stronger additive effect on starting weight, equivalent to 4.6% of the trait mean, was also detected on SSC6 (LIN3412 SNP probe). Interesting genetic effects on premium cuts yield were observed on the three chromosomal regions. An additive effect on ham weight of ASGA0019655 SNP probe (SSC4) was detected with a magnitude equivalent to a 1% of the mean. Two SNP probes (ALGA0037139 and H3GA0034325, on SSC6 and SSC12 respectively) showed significant additive effects on the loin weight, respectively equivalent to 4.4 and 2% of the mean. Moreover, a large dominant effect of the first probe on this trait was also estimated and was equivalent to a 7.4% of the mean. No significant SNP effects were identified on the composition of *longissimus dorsi* muscle.

Table 2. Significant additive (*a*) and dominant (*d*) effects on growth and carcass traits detected in the analysed chromosomal regions of SSC4, SSC6 and SSC12

Trait	SNP probe	SSC	a (SE)	d (SE)	LR*	d.f.†	P-value
Growth in fattening period							
Starting weight, kg	LIN3412	6	4.51 (1.23)	-	13.02	2	0.001
	ALGA0123491	12	3.11 (1.07)	-	8.23	2	0.016
Final weight, kg	ALGA0123491	12	2.14 (0.93)	2.91 (1.29)	9.98	3	0.019
Carcass weight, kg	ALGA0123491	12	2.10 (0.78)	2.01 (1.09)	10.04	3	0.018
Premium cuts yield‡							
Hams, kg	ASGA0019655	4	0.24 (0.09)	-	7.04	2	0.030
Loins, kg	ALGA0037139	6	0.13 (0.06)	-0.22 (0.07)	9.59	3	0.022
	H3GA0034325	12	0.06 (0.02)	-	7.06	2	0.029

*LR = Likelihood Ratio; † d.f. degrees of freedom; ‡ Adjusted for a common carcass weight.

The first question posed by the results concern the causal mutations underlying the observed genetics effects. Four probes (ALGA0123491, ASGA0019655, ALGA0037139 and H3GA0034325) correspond to SNPs located in intergenic regions, excluding ALGA0037139 that is an intronic mutation of *EFCAB7* gene. Only, the LIN3412 SNP probe corresponds to the sequence of a functional candidate gene. This SNP is an A/G substitution in the 3412 position of intron 11 of the *LEPR* gene sequence. Therefore, the potential functional effects of all these SNPs are not evident and their association with the phenotypic traits should be explained by linkage disequilibrium with the actual causal mutations.

It is interesting to compare the extent of LD in the *Iberian* population analysed with that observed in other pig breeds. Du *et al*. (2007) assessed the extent of LD in six lines of commercial pigs and found an average value of r^2 of 0.20 for SNPs separated by one cM. Despite the narrower distance between markers (0.34 cM), our results indicate a lower LD (r^2 = 0.10) in the *Iberian* population, which is a mixture of eight different *Iberian* breeding nuclei. A low LD is unfavourable to detect significant associations with quantitative traits, and more successful results may be expected within specific *Iberian* pig lines of lower effective size and greater LD.

A second relevant question is the usefulness of low-density SNP panels as tools for improving economically important traits of *Iberian* pigs. Breeding schemes for this breed are mainly focused on improvement of growth and carcass traits but quality traits should be also included in the breeding goal to take into account meat suitability for a long dry-curing process

(Fernandez *et al*., 2003). Meat quality and carcass performance traits are lately recorded on slaughtered pigs genetically related to the candidates to be selected as breeding animals. This reduces the intensity of selection and leads to a long generation interval. Methods for *in vivo* prediction of carcass and meat composition have been proposed for commercial pig breeds (Maignel *et al*., 2010) but, their application to heavy pigs as the *Iberian* may be difficult due to the differences in age, weight and body composition between tested candidates and commercial pigs. An alternative is the use of molecular markers associated to these traits. Our study explores an approach in this way combining results from QTL experiments and emerging high-throughput genotyping methods. The usefulness of this procedure will depend on its ability to provide early predictions of the genetic values for meat quality and carcass traits which would shorten the generation interval.

New high-throughput genotyping techniques are making a large amount of SNPs available that can be used to study patterns of genetic variation across the genome, to identify genome regions associated to traits of interest and to perform genetic selection with high accuracy. In particular, there is a growing interest in the application of genome wide selection (Meuwissen *et al*., 2001) that uses associations of large number of SNPs across the whole genome with phenotypes, without prior QTL detection. Genomic selection is the actual paradigm of animal breeding for traits difficult to improve by traditional methods. It could revolutionise *Iberian* pig breeding by producing early estimates of genetic merit of high accuracy for many more animals. However, some objections to this proposal may be outlined: (i) the instability of associations based on LD between neutral markers and traits limits its practical use to a reduced number of generations; (ii) many SNPs genotyped by the commercially available high-density porcine arrays are monomorphic in *Iberian* pigs, because this breed has not been taken into account in their design; and (iii) the development of high-density SNP arrays has excluded the information from QTL experiments and association studies performed in pigs along the last two decades; and (iv) finally, the high-density genotyping arrays have still a high cost for *Iberian* pig breeders.

Cheaper alternatives to genomic selection or genome wide association studies based on low-density SNPs panels have been suggested. Fontanesi *et al*. (2010) identified hundreds of SNPs in candidate genes for fat deposition traits from three different sources: sequencing, literature mining and expression database mining. To detect porcine genes affecting obesity related traits, they genotyped 677 selected SNPs in *Large White* pigs using a selective genotyping approach based on the estimated breeding value for back fat thickness (BFT). Eight genes have been associated with BFT, but possible SNPs effects on other traits related to fat metabolism have not been reported in their paper.

Our approach is also an alternative based on low-density SNPs panels but presents some singular features. The SNP identification was based on the results of previous QTL detection experiments which involved the *Iberian* breed as a parental breed, and the actual polymorphism of selected SNPs in *Iberian* pigs was verified using a moderate number of high-density genotyping arrays. The selective genotyping performed in the quoted study was exclusively based on one trait while we genotyped animals with many recorded traits for detecting marker-trait associations. Although this study is only a preliminary work, several interesting results have been obtained. Future research should focus in other chromosomal regions harbouring QTL for relevant traits and additional pigs with new records (e.g., fat thickness, back fat and muscle fatty acid composition or meat colour).

Acknowledgements

The authors wish to acknowledge the financial support received from the MICINN (Project AGL2007-65563-C02). Technical assistance of M.A. López, F. García, J. Salces, Y. Nuñez and A. Mercadé (UAB) and the facilities provided by E. de Pedro (UCO), J. García-Casco (INIA - Centro Nacional de Investigación en Cerdo Ibérico) and AECERIBER to obtain phenotypic records and samples are also recognized.

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Recovery program of Cádiz's Golden Iberian pig strain *(Dorado Gaditano)*

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Abstract. The diversity of the Iberian Pig, in last century, has suffered severe erosion due to the concentration of production in the most productive strains. This has led to the extinction of several Iberian pig breed subpopulations. One of them was Cádiz's Golden Iberian Pig Strain *(Dorado Gaditano)*, which was characterized by its golden glow hair. However, recently, *Dorado Gaditano* pigs were localized in different areas of the southern mountains of Cádiz province. These pigs are isolated and usually live wildy. This finding has ecological and cultural value. So members of several public institutions came together to promote a recovery program. This program was funded by a project in the 2008 call for *Conservation of Genetic Resources of Agrifood Interest* of the INIA. In this paper we expose the steps taken so far with a first recovery nucleus consisting of two boars, six sows and forty-five candidates for breeding animals (twenty-two males and twenty-three females).

Keywords. Cádiz Golden Iberian Pig Strain *(Dorado Gaditano)* – Feral domestic pigs – Wild biodiversity.

Programme de récupération de la variété de porc ibérique Doré de Cádiz (Dorado Gaditano)

Résumé. *L'isolement et la dérive génétique dans les populations de petite taille sont la principale source de variation intra-raciale. Le porc Ibérique a subi ce processus, accumulant au fil du temps une hétérogénéité génétique. Cependant, au siècle dernier, cette diversité génétique du porc Ibérique a été l'objet d'une importante érosion due à la concentration de la production dans les souches plus productives. Ceci a provoqué l'extinction de diverses sous-populations du porc Ibérique. L'une d'elles était le Dorado de Cádiz, souche qui se caractérisait par l'éclat doré de sa robe. Cependant, on a localisé récemment dans différentes régions des montagnes du sud de Cádiz, principalement dans les environs de Campo de Gibraltar, des porcs sauvages qui pourraient répondre à la morphologie du Dorado de Cádiz. Cette constatation a une incontestable valeur culturelle et écologique. Ainsi, les membres de diverses institutions publiques se sont réunis pour promouvoir un programme pour sa capture et sa récupération. Ce programme est financé par un projet de l'édition 2008 de la Conservation des Ressources Génétiques d'Intérêt Agroalimentaire de l'INIA. Dans cet article nous présentons les mesures prises jusqu'à ce jour, avec un noyau de récupération composé de deux verrats, trois reproductrices adultes et trente-deux candidats pour la reproduction (14 mâles et 18 femelles).*

Mots-clés. *Dorado de Cádiz – porcs sauvages – Biodiversité.*

I – Introduction

The Iberian Pig Breed is recognized as a heterogeneous breed characterized by its great intra-breed diversity including several lines and strains. This intra-breed diversity of the Iberian pig breed is usually structured on the basis of morphology and coat colour. The Govern of Spain has included five traditional Iberian pig strains in the Official Catalogue of Livestock Breeds of Spain (RD 2129/2008). Therefore the diversity of the Iberian Pig, in last century, has suffered severe erosion due to the concentration of production in the most productive strains. This has led to the extinction of several Iberian pig breed subpopulations. One of them was Cádiz's Golden Pig Strain (*Dorado Gaditano*), which was characterized by its golden glow hair and it was mainly located in mountains of Cádiz. This Iberian pig subpopulation are officially extinct since the nineties of last century.

However, possible Cádiz's Golden pigs were recently localized in different mountains of Cádiz province (southern Spain), mainly around the Campo de Gibraltar. These pigs live wildly as feral domestic pigs. They coexist with wild boars, other feral domestic pigs and crossbreed animals from wild boars x feral domestic pigs. This finding has ecological and cultural value. So some members of several public institutions came together to initiate a recovery program. This program is funded by a project approved in the 2008 call for *Conservation of Genetic Resources of Interest Agrifood* of the INIA. In this paper are described the steps taken to get the first recovery nucleus.

II – Materials and methods

1. Animal location, capture and sampling

Numerous locations of the mountains around of Campo de Gibraltar were explored to locate possible feral Cádiz´s golden Iberian pigs. The catches of animals were carried out with cages located in areas defined as capture zones. Inside these cages were placed baits (food). The captured animals were subjected to a preliminary morphological analysis to make an initial screening and determine their morphological adequacy. At the same time, blood samples from these captured animals were collected to carry out a preliminary cytogenetic study, genetic studies and health analyses for the notifiable diseases.

2. Morphological evaluation of the catches animals

All animals captured were morphologically evaluated according to the standard defined for *Dorado Gaditano* from previous morphological characterization studies (Mata *et al.,* 1998; Pardo *et al.,* 1998) and compiled old photos.

3. Cytogenetic and genetic studies

Captured animals were subjected to a preliminary cytogenetic study to reject crossbreeding with European wild boar and to avoid the main chromosomal defects. This study was based on the analysis of the karyotypes (chromosome number) and several chromosome bandeos. The karyotypes were prepared according to the recommendation of the Committee for the Standardized Karyotype of Domestic Pig (1988).

For the DNA molecular analysis, 34 microsatellite markers, including the 27 recommended by FAO for genotyping of pig populations (FAO, 2004), were used. Furthermore, the genotyping for SNP mutations of the IGF2 and MC1R genes were carried out using real time polymerase chain reaction (RT-PCR). This methodology differentiates Iberian pig breed from other breeds such as Duroc, lean breeds, etc. This methodology is officially used by AECERIBER to ensure racial purity of animals registered in studbook.

III – Results and discussion

After quarantine time, some of the captured animals were selected by their correct results for cytogenetic, molecular and serological analyses. These animals (two boars and three sows) were moved to the recovery nucleus as initial foundational population. This nucleus was located in the Experimental Agricultural and Livestock Center of the Provincial Deputation of Cádiz. In a short time, a secondary recovery nucleus (safety nucleus) is going to be located in the farm *"La Algaba"* in the *"Serranía de Ronda"*, due to its adequate isolation characteristics.

1. Genetic characterization studies

Regarding MC1R and IGF2 genes, all selected animals showed the characteristic genotype of the Iberian breed, ensuring the absence of introgression of foreign breeds.

In Table 1 are shown the main genetic variability parameters (Ho, He and alleles/locus) of then main Iberian pig subpopulation including the Cádiz´s Golden Iberian Pig Strain.

Table 1. Genetic diversity values by Iberian pig subpopulations

Subpopulations	He†	Ho†	Alleles / locus
Dorado Gaditano (DGA)	0.493	0.577	2.86
Entrepelado (ENT)	0.622	0.526	5.19
Torbiscal (TOR)	0.585	0.536	4.68
Negro Lampiño (NLA)	0.534	0.443	5.16
Manchado de Jabugo (MDJ)	0.319	0.352	2.26
Retinto (RET)	0.610	0.527	6.00
Alentejano (ALE)	0.559	0.608	4.23
Negro de los Pedroches (NDP)	0.605	0.616	4.10

†He: expected heterozygosity; Ho: observed heterozygosity.

As expected, the He value (0.493) and number of alleles per locus (2.86) for the *Dorado Gaditano* strain showed a worrying genetic diversity status in comparison with most of Iberian strains. This values are only exceeding those shown by the *Manchado de Jabugo*, an Iberian Pig Strain in serious danger of extinction with an estimated population size at around 30 individuals.

In Table 2 are shown F_{ST} values (a measure of genetic differentiation among subpopulations) between pairs of Iberian pig subpopulations. According to this F_{ST} values, *Dorado Gaditano* was genetically closer to *Retinto* and *Entrepelado* strains than to other analyzed subpopulations, while *Dorado Gaditano* was very genetically different to *Manchado de Jabugo.* Important levels of genetic differentiation were also observed between *Dorado Gaditano* and other Iberian pig strains such as *Torbiscal* and *Lampiño*.

Table 2. Genetic differentiation matrix (F_{ST}) between pairs of Iberian pig subpopulations

	Torbiscal	Lampiño	Alentejano	MDJ	NDP	Retinto	Dorado Gaditano
ENT	0.15	0.17	0.07	0.22	0.10	0.05	0.11
TOR		0.20	0.18	0.32	0.16	0.17	0.25
NLA			0.17	0.29	0.17	0.17	0.20
ALE				0.24	0.10	0.07	0.14
MDJ					0.28	0.22	0.45
NDP						0.10	0.17
RET							0.10

This genetic differentiation value is lower than genetic differentiation levels reported for other Iberian pig subpopulations in relation to the general Iberian pig breed population (Clemente *et al.,* 2008b). Also, this genetic differentiation value is much lower than the average value of genetic differentiation for the whole Iberian pig subpopulations, estimated at 0.190 (Clemente *et al.,* 2008a).

Furthermore, Reynolds genetic distance between *Dorado Gaditano* and several swine populations (different breeds and Spanish wild boar) were estimated from microsatellite allele frequencies (Table 3). The pig population for this analysis included several breeds such as Duroc, Creole (Mexican hairless pig), Mangaliça, commercial breeds (such as Large-White, Hampshire, Pietrain and Landrace), Iberian Pig (excluding *Dorado Gaditano* pigs) and Spanish wild boars.

Table 3. Reynolds genetic distance between swine populations (pig breeds and Spanish wild boar)

Reynolds genetic distances	Duroc	Creole	Sp. Wild boar	Mangaliça	Lean pig breeds	Dorado Gaditano
Iberian Pig	0.20	0.23	0.10	0.15	0.18	0.02
Duroc		0.28	0.27	0.34	0.27	0.28
Creole pigs			0.16	0.33	0.25	0.26
Sp. Wild boar				0.24	0.17	0.08
Mangaliça					0.25	0.27
Lean pig breeds						0.25

The extremely close genetic relationship between *Dorado Gaditano* and Iberian pig breed was outstanding. This confirms that *Dorado Gaditano* is genetically ascribed to the Iberian pig breed. The large genetic distances between *Dorado Gaditano* and each analyzed population (excluding Iberian breed and Spanish wild boars) are also noticeable.

Until recent decades, blond-haired Iberian pigs, such as *Dorado Gaditano* pigs (golden-haired pig), participated in livestock fairs within the red-haired varieties of Iberian breed (today grouped under the *Retinto* strain) without explicit differentiation among them. Therefore, there was usually gene-flow between blond and red Iberian pigs, which would be evidenced in this observed genetic proximity between *Dorado Gaditano* and *Retinto*. As result, blond varieties could be regarded as subpopulations derived from ancient red varieties group of Iberian breed (known today as *Retinto* in which are currently only included dark-red-haired pigs). On the other hand, *Lampiño* is the farthest subpopulation to *Dorado Gaditano*.

Finally, a conservation program was established alongside recovery program. One of the main preoccupations in this program is the containment of increase in inbreeding. So this conservation program includes a Studbook and a reproductive scheme between boars and sows assisted by morphologic characterization and genetic data, according to methodology described in Fernández *et al.* (2010). Actually, the recovery and conservation nucleus included two boars, six sows, and forty-five candidates for breeding animals (twenty-two males and twenty-three females).

IV – Conclusions

From a genetic standpoint, there is no doubt that the animals taken to the recovery nucleus are perfectly ascribed to Iberian pig breed. Similarly, these animals are a distinct group from other subpopulations of the Iberian pig breed, showing the specific characteristics that exhibited the extinct *Dorado Gaditano*. It should be noted that the capture zones coincide with areas where once was known the *Dorado Gaditano*. Therefore, the recovery of the extinct *Dorado Gaditano* strain, though not without difficulties, it seems possible.

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Genetic parameters and response to selection in several herds of Iberian pigs for piglets weight at 90 days of age

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Abstract. The Piglet Index is included in the official genetic scheme for the iberian pig breed, managed since 1992 by the Spanish Association of Iberian Pig Breeders (AECERIBER). This index allows the farmers to select the candidates according with the breeding value for piglet weight at early ages. The data analysed in this work came from 14 breeds with, at least, 1000 piglets controlled in their weight in 10 different seasonal batches. First, heritabilities (h^2) and litter effect coefficient (c^2) estimates were calculated in each breed using an animal model with fixed and random effects. After, the regression of the average breeding value along time (genetic trend) was estimated also in all 14 breeds. Heritabilities and litter coefficients estimates are quite heterogeneous in their values. So, h^2 ranges from 0.00 to 0.24 and c^2 from 0.15 to 0.64. In eight breeds h^2 reaches values next to zero, results that stress the importance of the quality information (genealogic and productive) provided by the breeder. Genetic trend were positive and significant in eight breeds.

Keywords. Piglet index – Heritability – Genetic trend.

Paramètres génétiques et réponse à la sélection dans plusieurs troupeaux de porcs Ibériques pour le poids des porcelets à 90 jours d'âge

Résumé. *L'index concernant les porcelets est inclus dans le schéma génétique officiel pour la race Ibérique de porc, contrôlé depuis 1992 par l'association espagnole des sélectionneurs de porc Ibérique (AECERIBER). Cet index permet aux éleveurs de choisir les candidats selon la valeur génétique d'amélioration pour le poids des porcelets à un âge précoce. Les données analysées dans ce travail proviennent de 14 élevages avec, au moins, 1000 porcelets contrôlés pour leur poids dans 10 différentes parités saisonnières. D'abord, des estimations d'héritabilité (h^2) et de coefficient d'effet de la portée (c^2) ont été calculées dans chaque élevage en utilisant un modèle animal avec des effets fixes et aléatoires. Après, la régression de la valeur génétique d'amélioration moyenne sur le temps (tendance génétique) a été estimée également dans chacun des 14 élevages. Les estimations d'héritabilités et de coefficients de portée sont tout à fait hétérogènes quant à leurs valeurs. Ainsi, h^2 s'étend de 0,000 à 0,243 et c^2 de 0,153 à 0,640. Dans huit races h^2 atteint des valeurs proches de zéro, résultats qui soulignent l'importance de la qualité de l'information (généalogique et productive) fournie par le sélectionneur. La tendance génétique était positive et significative dans huit élevages.*

Mots-clés. *Index des porcelets – Héritabilité – Tendance génétique.*

I – Introduction

In 1992 the Ministry of Agriculture approved the Official Genetic Improvement Scheme for the Iberian Pig Breed (BOE 1992). Since the beginning of the selection program the Spanish Association of Iberian Pig Breeders has managed this Scheme. Besides a genetic index for finishing pigs (the so call Complete Cycle Index, AECERIBER 1998), several breeds take part in the scheme evaluating piglets for weight at early age (an intraherd Piglet Index, AECERIBER, 1998). The present work is a summary of the activity carried out for 14 herds with more than

1000 piglets controlled in their weights, distributed in, at least, 10 batches (group of piglets born in the same year-season). In order to preserve the identity of the herds involved in this article, they are named as H1-H14.

II – Herds and data

Table 1 gives details of the information related with the 14 herds included in this study.

The quantity and quality of the information in each herd is quite unequal; so are their sizes. As example, some of them weigh piglets once a year (H12 with 12 batches controlled in 11 years) while others, as H8, has records nearly in all the piglets born in the herd.

Table 1. Number of piglets controlled, batches and period were they took place, litters, breeding animals parents of the piglets, animals in the pedigree and, finally, sows registered in the Herd Book of the breed (census herd) in the 14 herds

Herd	Piglets controlled	Batches	Period (year-season)	Litters	Boars	Sows	Pedigree	Census
H1	1093	17	spr04-aut09	139	13	80	1132	66
H2	1890	20	aut99 - aut09	365	17	83	1928	31
H3	3382	24	spr04-aut09	544	12	125	3453	43
H4	1068	13	spr04-aut09	219	18	118	1153	113
H5	3009	27	aut94-aut09	492	23	110	3067	26
H6	2900	24	aut94-spr09	528	38	187	3266	107
H7	1930	14	aut99-spr09	310	28	229	2330	190
H8	7138	45	aut00-sum09	1188	18	369	7859	162
H9	1887	15	spr04-aut09	297	19	187	2039	147
H10	2242	15	aut01-aut09	324	20	136	2390	78
H11	2147	11	spr02-spr08	345	25	155	2264	58
H12	1289	12	aut99-aut09	195	14	128	1413	80
H13	2485	14	win01-spr07	453	35	153	2668	49
H14	4233	25	sum91-aut07	731	57	480	4502	69

Table 2 contains the main statistics in the 14 herds. The final value of the trait was the weight at the exact ages indicate in the Table obtained after a correction based in the daily gain between two consecutive weights.

Only H3, H4 and H9 work on the piglet weight at ages earlier than the rest (65-70 days), which is reflected in an inferior average weight, around 20 kg. The trait can be considered like of moderate variability, with coefficient of variation located between 17% (H10) and 27%. (H6, H7 or H9). It is important to emphasize that there are neither genealogic nor productive connections between the herds, except for some boars purchased among them. As the herds have specific conditions of climate, geographic location, facilities and handling, feeding, etc, the averages of Table 2 cannot be compared.

III – Analysis and results

1. Genetic parameters

In the first place an analysis of components of variance in each one of the herd has been carried out, with the purpose of calculate the genetic and environmental parameters. In Table 3 the obtained results appear, obtained by maximum likelihood with a linear model that includes sex and batch as fixed effects and animal and litter as random effects.

Table 2. Ages of weight, averages, standard deviations and coefficients of variation in the 14 herds

Herd	Age (days)	Average weight (kg)	Standard deviation (kg)	Variation coefficient
H1	90	29.9	5.4	18.1
H2	90	26.6	6.2	23.3
H3	70	20.4	3.9	19.3
H4	65	21.6	5.6	25.8
H5	90	27.8	5.2	18.6
H6	90	25.3	6.8	26.9
H7	90	25.0	6.9	27.4
H8	90	26.1	5.2	19.8
H9	70	19.6	5.3	27.3
H10	90	30.4	5.2	17.0
H11	90	24.3	5.2	21.6
H12	90	26.3	6.1	23.4
H13	90	24.0	5.4	22.3
H14	90	25.0	6.2	24.9

Table 3. Variance components (Va, Vc, Ve y Vp), heritabilities and litter coefficients in the 14 herds

Herd	Variance components				Heritability	Litter coefficient
	Vp	Va	Vc	Ve		
H1	8.30	0.08	1.27	6.95	0.01	0.15
H2	16.68	3.19	5.91	7.58	0.19	0.35
H3	7.75	0.38	4.96	2.40	0.05	0.64
H4	21.53	1.19	8.21	12.14	0.06	0.38
H5	22.20	1.16	9.38	11.66	0.05	0.42
H6	23.24	1.76	10.06	11.42	0.08	0.43
H7	11.64	0.00	3.81	7.83	0.00	0.33
H8	14.81	3.33	2.55	8.92	0.23	0.17
H9	14.22	3.30	5.03	5.90	0.23	0.35
H10	19.80	4.20	4.26	11.35	0.21	0.22
H11	23.56	1.66	9.42	12.49	0.07	0.40
H12	29.84	7.24	13.57	9.03	0.24	0.46
H13	24.51	1.62	7.84	15.06	0.07	0.32
H14	23.54	2.62	7.56	13.36	0.11	0.32

The results are very different among herds, as much in the variance components (differences in the total variance very great in H1 and H3 as opposed to H12), as in the coefficients. Whereas in five herds (H2, H8, H9, H10 and H12) estimates of h^2 present values next to those found in other similar studies (between 0.20 and 0.25) in Iberian pig or in white coat races (NSIF, 2002) which allows to suppose significant answers to the selection, in other eight herds (H1, H3, H4, H5, H6, H7, H11 and H13) these values of h^2 are very low, less than 10%. In a last herd (H14), heritability reaches an intermediate value between the other two groups.

The result obtained in H7, since the additive genetic variance is zero ($h^2 = 0$), and in H1 where

h^2 remains in 0.01, is worthy of mention. In the Iberian pig, even though the handlings and facilities in many farms are remote still of the habitual ones in intensive pig production, in which the environmental factors are controlled and are more homogenous, it is not expectable that the contribution of these environmental factors to the total variance of the character (values of Ve and Vc) is so important in magnitude so as that the coefficient of the genetic variance (that is, the heritabillity) is practically null. Therefore, these values of h^2, almost null, must be related to the quality of the information that from the herd has been transmitted to carry out the genetic evaluations, or because the genealogies are not the correct ones, or because abundant errors in the weights taken in the pigs exist.

The effect of the common environment that the pigs born in the same litter share, is very important in these traits measured in so early ages. Apart from three herds, again H1, H8 and H12, c^2 always surpasses 30%. Possibly in camping farrowing systems these environmental factors have greater importance. However, in H3 c^2 reached 64%, value that seems excessive for the weight to 70 days.

2. Genetic trends

The average of the genetic evaluations of all the piglets born in the same batch were obtained using the parameters of Table 3. Later, the value of the regression over the time of this average was calculated, as an estimate of genetic trend. Table 4 and the Figure 1 (herds H2 and H8) show these results.

Table 4. Coefficient of determination (R^2) and of regression (b), probability associated with b=0, cumulative response, percentage of the response over the mean trait ant economic response per piglet in the 14 herds

Herds	R^2	*b* (kg)	Prob>T	Cumulative response (kg)	Percentage over the mean trait	Economic response per piglet (€)
H1	0.39	0.003	0.0070	0.04	0.15	0.02
H2	0.96	0.251	0.0001	5.02	18.87	1.94
H3	0.08	0.002	0.1819	0.05	0.26	0.02
H4	0.07	0.010	0.3831	0.13	0.61	0.05
H5	0.70	0.028	0.0001	0.76	2.72	0.29
H6	0.72	0.038	0.0001	0.90	3.56	0.35
H7	0.21	0.000	0.1004	0.00	0.00	0.00
H8	0.64	0.036	0.0001	1.62	6.19	0.63
H9	0.11	0.037	0.2327	0.56	2.87	0.22
H10	0.66	0.080	0.0002	1.20	3.93	0.46
H11	0.40	0.053	0.0374	0.59	2.41	0.23
H12	0.20	0.098	0.1423	1.17	4.47	0.45
H13	0.33	0.031	0.0196	0.43	1.81	0.17
H14	0.18	0.026	0.0356	0.65	2.58	0.25

The results of Table 4 show a positive and significant genetic response in herds H2, H5, H6, H8, H10, H11, H13 and H14. In four of them (H2, H8, H10 and H14) estimates of h^2 were elevated and the suitable selection of the breeding animals has allowed this good outcomes. In other two (H5 and H6) in spite of the low value of h^2 (0.052 and 0.076, respectively) their perseverance in the participation of the Scheme and the selection carried out throughout this period has been translated also in positive responses (around 3% of the trait mean). The genetic trend in herds H9 and H12 indicates values similar to the previous ones, but the adjustment to the regression is very weak, non significant, due to great fluctuations that move away much of the regression straight line. In the four remaining herds (H1, H3, H4 and H7), both because of the low heritability as of the own selection of breeding animals, the results pointed to absence of response.

Some comments needs the results of H2, whose policy of selection of boars and sows is totally conditioned by the breeding values of the candidates. It allows an accumulated response of 29% of the trait mean (Figure 1). The approximated economic response would be of almost 2 € per piglet maintained in the farm until 90 days of age. Figure 2 corresponds to the phenotypic trend in H2.

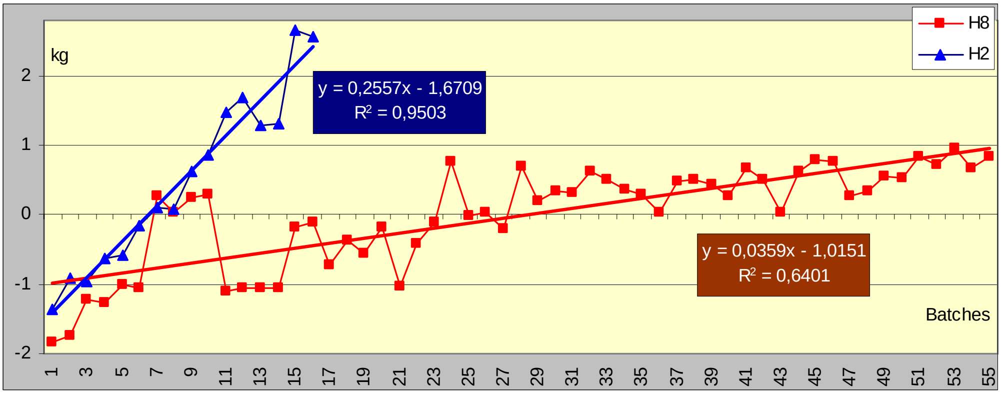

Fig. 1. Graphical representation of the average of the genetic evaluations of the animals born in each batch throughout time in the herds G8 and G2, with the line and the equation of the regression with its coefficient of determination, obtained in the analysis with the parameters of Table 3.

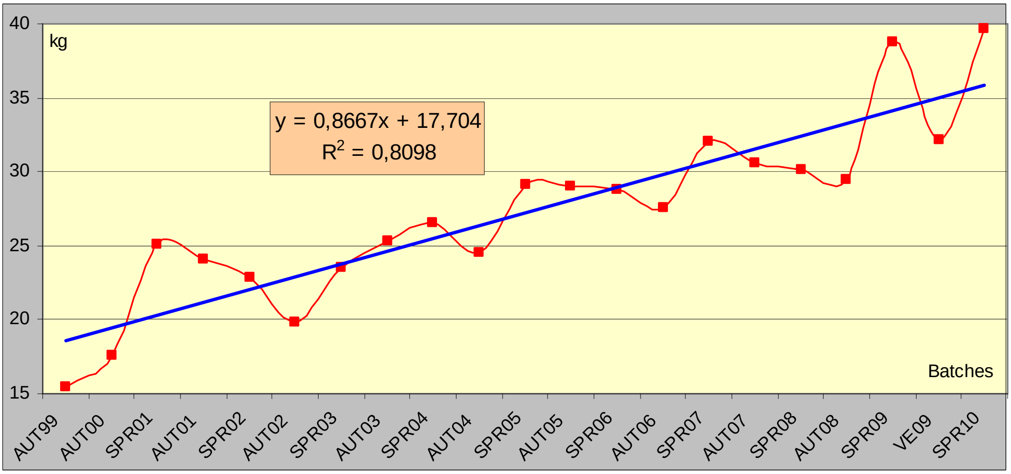

Fig. 2. Graphical representation of the phenotypic trend in the herds H2, with the line and the equation of the regression with its coefficient of determination.

IV – Conclusions

The present study allows drawing some lessons:

(i) It is possible to obtain important genetic improvements (like in herds H2, but also in H5, H6, H8, H10 or H12) in few years in certain characters of medium heritability, through the suitable election of the reproducers, especially of boars. Growth (average daily gain) and carcass composition traits (percentage of premium cuts), with greater economic relevance than piglet weight at early ages and that fulfils with the premise about h^2 magnitude, are excellent candidates for their selection in fattened Iberian pure pigs.

(ii) The genetic evaluation by itself, if it does not have any incidence in the selection of the new boars and sows, constitutes an extra effort without positive effects for the herd.

(iii) The quality of the productive and genealogical information provided by the herds to make possible the genetic evaluations is essential, since it affects to the correct estimation of the genetic parameters (heritability) and to the expected response to selection (possibility of erroneous selection of the breeding animals).

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Genetic parameters and trends for litter size in Black Slavonian pigs

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Abstract. Genetic progress in litter size in local pig breeds is limited mainly due to low heritability and inbreeding depression. After increase of population size of Black Slavonian (BS) pig breed in recent years, there is opportunity to renew a breeding programme including some new breeding goals related to litter size. Aim of study was to determine genetic parameters for litter size in BS pigs. Litter size was presented as number of piglets born alive (NBA) and number of piglets weaned (NW). Genetic parameters were estimated using a univariate repeatability models. Fixed part of the model included breeding organization, birth number, farrowing season and service sire as class effects. Random part of the model included direct additive genetic effect and permanent environmental effect. Litter size records proceeded from 5792 litters from 1817 sows. The complete pedigree file contained 1862 triads (animal-sire-dam). Heritability estimates for NBA and NW were lower than average values of 0.10 - 0.20, and ranged for both traits between 0.04 and 0.08. Implementation of some others models in breeding scheme (particularly multivariate and random regression models) requires availability of large set of reproductive traits for this local breed.

Keywords. Pigs – Local breed – Genetic parameters – Litter size.

Paramètres génétiques et tendances de la taille de la portée chez le porc slavonien noir

Résumé. *Le progrès génétique de la taille de la portée chez les races locales est limité essentiellement en raison de la faible héritabilité et de la dépression de consanguinité. Après l'augmentation de la taille de la population du porc slavonien noir (SN) au cours des dernières années, il est maintenant possible de renouveler les programmes d'amélioration en y incluant de nouveaux objectifs relatifs à la taille de la portée. L'étude avait pour but de déterminer les paramètres génétiques de la taille de la portée des porcs SN. Cette taille a été présentée sous la forme du nombre de porcelets nés vivants (NNV) et du nombre de porcelets sevrés (NS). Les paramètres génétiques ont été estimés en utilisant le modèle univarié de répétabilité. La partie fixe du modèle est composée de l'organisation de l'amélioration, du numéro de mise bas, de la saison de la mise bas, du reproducteur mâle comme effets liés à la classe. La partie aléatoire du modèle est composée de l'effet génétique additif direct et de l'effet environnemental permanent. Les dossiers sur la taille de la portée sont basés sur l'analyse de 5792 portées de 1817 truies. Le dossier complet des pedigrees contenait 1862 triades (animal-père-mère). Les estimations d'héritabilité pour les NNV et les NS sont inférieures aux valeurs moyennes de 0,10-0,20, et varient pour les deux caractères entre 0,04 et 0,08. La mise en œuvre d'autres modèles dans le schéma d'amélioration (en particulier des modèles multivariés et de régression aléatoire) exige la disponibilité d'un grand nombre de caractéristiques reproductives pour cette race locale.*

Mots-clés. *Porcs – Race locale – Paramètres génétiques –Taille de la portée.*

I – Introduction

The Black Slavonian (BS) pig is one of two autochthonous pig breeds in Croatia. The breed was developed at the end of the 19th century in the east part of Croatia (Slavonia region). Until middle of 20th century it was the most widespread breed in the country. A period of extinction of these breed lasted until middle 1990s, when number of pigs was reduced to only 60 sows and 5 boars (Uremovic *et al*., 2001). Since 1996 population of BS pigs increased, primarily due to state protection measures and individual efforts of pig breeders. The breed is kept traditionally in outdoor system and meat from BS pigs is used mainly for

production of local meat products (Karolyi *et al*., 2007). In the last few years, population of BS pigs increased to more than 600 sows and 70 boars. The increase of population size opens a possibility to renew the breeding programme of this local breed.
Litter size is one of the economical most important traits in pig production. Litter size of BS pigs is low compared to modern pig genotypes, on average 6 to 7 piglets born alive (Sencic *et al*., 2001). Increase of litter size of BS pigs is crucial for production of sufficient amounts of local meat delicious, like Slavonian kulen sausage). The objective of this study was to determine genetic parameters for litter size of BS pigs and to consider the possibility to increase litter size by selection.

II – Materials and methods

Data were supplied by the Croatian Agricultural Agency. Litter size were presented as number of piglets born alive (NBA) and number of piglets weaned at 21 days (NW). Litter records were collected from sows farrowed between January 1998 and December 2009. In total there were 5792 litter records from 1817 sows (Table 1). Data file contained only litters from the first to the tenth parity. The complete pedigree file contained 1862 triads (animal-sire-dam).

Table 1. Basic statistics for number of piglets born alive (NBA) and number of piglets weaned (NW)

Trait	Min	Max	Mean	SD
NBA	1.0	16.0	6.14	1.75
NW	0.0	15.0	4.76	2.60

Genetic and environmental parameters were analyzed using a univariate repeatability model assuming complete genetic correlations between successive parities, for each trait separately. Fixed part of the model included breeding organization, parity, farrowing season, and service sire effect. Litter records were obtained form eight breeding organization, while farrowing season was defined as month-year interaction. In the twelve year period piglets from litters were descendants from 241 different service boars. The random part of the model included direct additive genetic effect, and permanent environmental effect. Computations were performed using the VCE6 software (Groeneveld *et al*., 2008).

III – Results and discussion

The estimated values of variances and heritabilities for NBA and NW (Table 2) of BS pigs obtained using univariate repeatability models were substantially lower than estimates in genetic analysis of modern pig genotypes with heritabilities ranging mainly between 0.10 and 0.15. Particularly low values of permanent environmental variances indicate possible bad data structure. Phenotypic variance for NW was doubled in comparison to NBA as consequence of a similar increase of the residual error variance. Some similar values of heritabilities for NBA in this study were found in some studies on Iberian pigs (Pérez-Enciso and Gianola, 1992; Fernández *et al*. 2008). Fernández *et al*. (2007) suggested that multiple-trait models should be recommended to estimate breeding values for litter size in Iberian pigs when the first and later parity records are involved. Further, they noted that a model for litter size treating NBA records at the first parity as one trait, and records at the later parities as second trait could be more advisable than the repeatability model.

Table 2. Variance† and heritability (h^2) estimates for number of piglets born alive (NBA) and number of piglets weaned (NW) under the univariate repeatability models

Trait	Var(a)	Var(p)	Var(e)	Var(ph)	h^2
NBA	0.21	0.02	2.27	2.50	0.08
NW	0.22	0.01	5.50	5.73	0.04

†Var(a): direct additive genetic variance; Var(p): permanent environmental variance; Var(e): residual error variance; Var(ph): phenotypic variance.

IV – Conclusions

The values of variances and heritabilites obtained in our analysis using univariate repeatability model were not promising too much for selection purposes. Some other strategies for estimation of genetic and environmental parameters, as multivariate approach and random regression models must be taken into consideration in the future. Additional attention must be also given to data quality, especially to the control of pedigree precision and registration of reproductive data.

Acknowledgements

The authors wish to acknowledge the financial support received from Ministry of Agriculture, Fisheries and Rural Developments (Project: Prediction of breeding values of pigs from family farms).

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Genetic structure of Krškopolje pig based on pedigree data

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Abstract. Krškopolje pig was persecuted for many years and consequently, population extremely decreased. In the year 1992 when the herdbook was established again, there were less than 30 sows and only 3 boars. Nowadays, the population consists of 150 registered sows and 30 boars. Pedigree data included 730 animals: 162 males and 568 females. Animals born in years 2005-2009 were considered as reference population. The PEDIG program was used for pedigree analyses. Generation intervals were 2 years for sire-son and sire-daughter, and 2.5 years for dam-son and dam-daughter pathways. Average family size for pairs sire-dam was 1.67 progeny with standard deviation (SD) 1.18, while sires as families had 4.08 progeny (SD 5.54). Maximum number of generations was nine. Pedigree completeness was expressed by equivalent complete generations: 3.2 in males and 3.3 in females. Total number of founders was 58. Effective number of founders and ancestors were 20 and 14, respectively. Average coefficient of kinship was 4.9% among males, 4.6% among females and males, and 5.0% among females. Due to incomplete pedigree, coefficients of inbreeding and relationship are underestimated, while effective number of founders and ancestors are overestimated. Sires should have more uniform contribution to the gene pool.

Keywords. Krškopolje pig – Endangered breed – Pedigree analysis – Genetic variability.

La structure génétique du porc Kraškopolje basée sur les données de son pedigree

Résumé. *Le cochon de race Kraskopolje a été persécuté pendant de nombreuses années ce qui a provoqué une forte diminution de sa population. À partir de 1992, lorsque le livre généalogique a été remis en place, il ne restait plus que 30 truies et seulement 3 verrats. De nos jours, la population est représentée par 150 truies et 30 verrats enregistrés. Les données concernant le pedigree sont basées sur 730 animaux: 162 mâles et 568 femelles. Les animaux nés durant les années 2005 à 2009 sont considérés comme la population de référence. Le programme PEDIG a été utilisé pour les analyses de pedigree. L'intervalle choisi entre 2 générations est de 2 ans pour la descendance père-fils et père-fille et de 2 ans et demi pour la descendance mère-fils et mère-fille. La taille moyenne d'une famille père-mère est de 1,67 descendants avec un écart-type de 1,18 ; alors que le père peut avoir plusieurs familles et au total 4,08 descendants (avec un écart-type de 5,54). Le nombre maximum de générations est quant à lui de 9. La profondeur du pedigree est, en équivalent de générations complètes, de 3,2 pour les mâles et de 3,3 pour les femelles. Le nombre total de fondateurs est de 58. Le nombre de fondateurs efficaces et d'ancêtres est respectivement de 20 et 14. Le coefficient de parenté a été estimé à 4,9% entre les mâles, à 4,6% entre les mâles et les femelles et à 5% entre les femelles. Le coefficient de consanguinité a été sous-estimé à cause d'un pedigree incomplet et d'un nombre d'animaux fondateurs et d'ancêtres qui a été surestimé. La contribution génétique des mâles devrait être plus uniforme.*

Mots-clés. *Porc Kraškopolje – Race en voie d'extinction – Analyse de pedigree – Variabilité génétique.*

I – Introduction

The Krškopolje pig or black-belted pig is the only preserved Slovenian indigenous pig breed. It originates from the south-east part of Dolenjska region, the area of Krško-Brežiško field and in the foothills of Gorjanci hills. The breed is adapted to poor rearing environment. It has large appetite, great ability to produce fat, good meat quality, good resistance, good maternal traits, and moderate fertility traits. The first known written record about Krškopolje pig dates from the middle of 19th century. Rohrman (1899) a prepared detailed description of the breed and he also named the breed as Krškopolje pig. He stressed that pigs in the Krško field are uniform in exterior, color, and also in other traits, and they are distinguishable from other breeds. However, many breeders from that time mixed and crossed the Krškopolje pig with Yorkshire. In the rural survey before the Second World War, Oblak (1938) pointed out that original Krškopolje pig could be found only it the most distant villages. The breed was prosecuted in the past and only the most persistent breeders deserve that breed still exists. The breed was put to the list of endangered Slovenian breeds in the year 1991. The herdbook was established in the year 1992, three family farms were included in gene bank and they started with pedigree and performance recording. At that time, population consisted of less than 30 sows and only 3 boars. After the year 2003, the interest of raising the Krškopolje pigs has increased. Consequently, the breeding herd in the year 2009 consisted of 104 sows and 24 boars.

Nowadays, the analysis of pedigree structure in populations of farm animals are more and more useful and uses tools that allows an overview of genetic background and development of population. It enables the assessment of genetic diversity in population, contribution of founders and ancestors, as well as contribution of genes from foreign populations. The results could be used for gradual change in endangered populations through the balanced contributions of ancestors to the gene pool, uniform family size and use of large number of unrelated sires. The purpose of the study was estimation of genetic diversity in the Krškopolje breed based on the pedigree data by different measures of genetic variability in the population.

II – Material and methods

The pedigree data of Krškopolje breed was provided by Central breeding organization for pigs. Data set consisted of animal identification, gender, sire, dam, birth date, origin, owner, and culling date. Animals born in years 2005 - 2009 were considered as the reference population.

Description of population comprises two groups of parameters, the first one is demographic and the second is genetic description. Demographic description includes number of males and females in population, their changes over time, generation interval, and family size. Genetic description which bases on probabilities of gene origin covers pedigree completeness, inbreeding coefficient and coefficient of kinship, contribution of ancestors and founders, equivalent number of known generations (Maignel *et al*., 1996), effective number of founders and ancestors (Boichard *et al*., 1997). The average relatedness (Dunner *et al*., 1998) was computed, as well. It enables choice of animals with smaller contribution and consequently slower increase of inbreeding in population, as well as slower losing of alleles from the gene pool of population. The PEDIG programme package (Boichard, 2002) was used for pedigree analyses.

III – Results and discussion

Demographic description. Altogether, pedigree data comprised of 730 animals (Table 1). The pedigree data in Krškopolje pig is collected for last 19 years, since herdbook was established in 1992. The proportion of animals with unknown parents was 7.94%. The reference population – animals born between years 2005 and 2009 – included 390 animals, 80 boars and 310 sows. There were 47 sires and 121 dams. Ratio between dams and sires was suitable (2.57) in reference population. All animals born in last years have both parents known.

Generation interval. Only progeny which had their own progeny from complete pedigree were included in computation of generation interval. Sires had sons and daughters when they were 2 years old (Table 2). Dams were on average half year older when they had offspring: 2.46 years at birth of sons and 2.65 years at birth of daughters.

Family size. More than family size, the uniformity of family size is important, because it affects maintenance of genetic variability and long-term survival of a population. The family size is presented by number of progeny per pair sire-dam, per sire and per dam (Table 3). The majority of pairs had only one progeny (61.0%). On average, pairs had 1.67 progeny with standard deviation of 1.18. Boars as families had on average 4.08 offspring with large standard deviation (5.54). Dams had on average less progeny compared to sires, as expected. The average number is 2.03. The variability for dams is also smaller, standard deviation was 1.35.

Table 1. Demographic description of complete pedigree and reference population

Item	Complete pedigree	Reference population
Number	730	390
Boars (Sires)	162	80 (47)
Sows (Dams)	568	310 (121)
Ratio dam / sire		2.57
Proportion of founders (%)	7.95	0.00

Table 2. Generation interval by gender of parents and offspring*

Path	No. of parents	No. of offspring	Generation interval (yr.)
Sire – son	16	37	1.99
Sire – daughter	22	97	2.01
Dam – son	28	39	2.46
Dam – daughter	67	124	2.65

*Only offspring with their own progeny are accounted.

Table 3. Family size*

Family	No.	Avg.	SD	Max.	D1 (%)**
Sire – dam	123	1.67	1.18	8	61.0
Sire	51	4.08	5.54	34	37.2
Dam	102	2.03	1.35	8	45.1

*Only offspring with their own progeny are accounted.
**Proportion of families with only one progeny

Table 4. Inbreeding coefficients

Class	Number	(%)
$0.00 \leq x < 0.05$	139	83.7
$0.05 \leq x < 0.10$	26	15.7
$0.10 \leq x < 0.15$	1	0.6
Nº. of inbred animals	166	22.7
Avg. inbr.	0.024	
Max. inbr.	0.125	

Inbreeding and kinship. Small proportion of inbred animals (22.7%) is consequence of incomplete pedigree (Table 4). The average inbreeding coefficient is underestimated, too. From the year 1992, when identification of Krškopolje pigs was started again, matings of close related animals were avoided and recently, for each farmer's sows the boar is chosen on the basis of smallest relatedness. Coefficient of inbreeding show us, how ancestors of an animal were related, however it does not tell us the relationship of an animal to another one. Coefficients of relationship were calculated among boars, among sows and between pairs boar-sow (Table 5). Average values are between 0.045 and 0.050, but they are most probable underestimated, too.

Table 5. Coefficient of relationship in the reference population

Pair	No. of pairs	Avg.	SD	Max.
Boar – Boar	3160	0.049	0.050	0.278
Boar – Sow	24800	0.046	0.045	0.307
Sow – Sow	47895	0.050	0.045	0.303

Pedigree completeness. Animals born in years 2005-2009 had at most 9 generation of ancestors known (Table 6). Completeness of pedigree is showed by the equivalent number of known generations. In boars, value for this parameter was 3.18 and for sows 3.29. On average animals in the reference population had 26.6 (boars) and 28.5 (sows) known ancestors.

Table 6. Equivalent number of known generations, average number of known ancestors, contribution of founders and ancestors for boars and sows in the reference population

Item	Boars	Sows	Item	For boars	For sows
Number	80	310	No. of founders	37	40
Max. no. of generations i	9	9	Effective no. of founders	20.6	19.7
Equivalent no. of generations	3.18	3.29	Effective no. of ancestors	13.9	14.0
Avg. no. of known ancestors	26.6	28.5	$N_{50\%}$*	5	5
			C_{max} (%)**	15.2	14.2

*Number of ancestors which contribute to gene pool 50%.
**The largest marginal contribution of single ancestor to gene pool.

Contribution of ancestors. The reference population had around 40 ancestors. The effective number of founders was around 20 and effective number of ancestors was 14. The smaller effective number of ancestors is expected, as showed Boichard *et al*. (1997). Both parameters are overestimated due to incomplete pedigree. Maximal marginal contribution of single ancestor to gene pool was 15.2% in boars and 14.2% in sows; only 5 ancestors contribute 50% to gene pool.

IV – Conclusions

The Krškopolje breed is an endangered population. It was enlarged in recent years; also the number of farmers who wish to raise this breed had increased. Compared to modern pig breeds, it is less productive, but the meat and dried products are tastier. Due to incomplete pedigree, parameters of genetic variability are underestimated (inbreeding, relationship) or overestimated (effective number of founders and ancestors). Contribution of individual animals to next generation should be equalised to preserve the breed for long-term.

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Differences in physico-chemical composition of loin of three sires from Duroc x Iberian cross selected according to their conformation

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Abstract. In the study we are going to determine the influence of animal selection taking into account its anatomical conformation in relation to the physico-chemical composition and the colour of the loin from the offspring of three Duroc (DU) boars with different anatomical conformation, and Iberian sows. Thus, 29 loins from the barrows of the offspring of DU x Iberian were analyzed. All animals were reared on the same farm in intensive regimen and fed with the same type of feed. The loin and ham pH were determined 45 minutes and 24 hours postmortem. We also determined the pressure-induced weight loss, water activity, the colour (L, a* y b*) and the chemical composition (moisture, fat, protein and ashes) of the *Longissimus dorsi* muscle. Significant differences were found in the three pig sires studied for the content of protein and fat. In this way, the samples from the least conformed sire had the highest fat content, whereas the samples from the most conformed sire had the most protein content.

Keywords. Duroc x Iberian – Anatomical conformation – Physico-chemical composition – Colour.

Différences de composition physico-chimique de la longe de trois lignées croisées Duroc x Ibérique sélectionnées en fonction de leur conformation

Résumé. *L'objectif de cette étude consistait à déterminer l'influence de la sélection animale pour les paramètres productifs de la première lignée obtenue de trois mâles de race Duroc (DU) et trois femelles Ibériques sélectionnées selon leur conformation anatomique. 29 longes de mâles castrés du croisement DUxIbérique ont été analysées. Tous les animaux ont été élevés dans la même ferme avec un régime intensif et nourris avec le même type de tourteau. Le pH de la longe et du jambon, à 45 minutes et 24 heures post mortem, les pertes de poids par pression, l'activité de l'eau, la couleur (L, a* et b*) et la composition chimique (humidité, matière grasse, protéines et cendres) du muscle* Longissimus dorsi*, ont été déterminés. Dans les trois lignées on a trouvé des différences significatives pour la teneur en matière grasse et en protéines. Les échantillons appartenant à la lignée la moins bien conformée ont montré la teneur la plus élevée en graisse, tandis que les échantillons appartenant à la lignée mieux conformée ont montré la teneur en protéines la plus élevée.*

Mots-clés. *Duroc x Ibérique – Conformation anatomique – Composition physico-chimique – Couleur.*

I – Introduction

The Iberian pig breed has recently increased in popularity because consumers value the high quality of the meat and meat products. The main characteristic that differentiates the Iberian pig from other breeds of white pig is its higher intramuscular fat content. Various studies have demonstrated that the meat from the Iberian pig is of higher quality than that which is obtained from industrial pigs (Estévez et al., 2003).

However, the Iberian pig has a slow growth rate as well as a low reproductive rate. Thus, in order to increase its productivity, it is frequently crossbred with the Duroc white pig at a level of 50% to 75% (B.O.E., 3/11/2007). This raises its reproductive capability, improves the growth rate and increases alimentary efficiency and lean fat content (Dobao et al., 1986) without a significant reduction in either the quality of the meat, meat products (López-Bote, 1998) or the parts of the pig that are highly regarded and considered of higher quality than others (García-Macías et al., 1996). In general, the genetic lines of the Duroc and its crosses yield carcasses with a higher fat content than the conventional white line, depending on the selection line of the Duroc, the percentage of Duroc genes in the final product and the line with which it is crossed.

The overall objective of this study was therefore to determine the quality of pork loin of the genetic cross between Duroc and Iberian pigs according to the anatomical configuration by means of the determination of the physical-chemical composition and the colour.

II – Materials and methods

1. Animals

Twenty-nine pigs bred from pure Iberian females and a varied group of Duroc males were slaughtered. All of the animals were castrated, reared on the same farm and followed a strict diet consisting of the same type of feed that was changed based on the stage of growth.

2. Samples

The muscle Longissimus dorsi (loin) was taken from the carcass. The samples were then placed into three categories depending on the anatomical configuration of the male Duroc progenitor.

To make a physical-chemical analysis, the sample was triturated with a mincer until a homogenous mass was obtained. This mass was preserved in hermetically sealed jars at a temperature of -20ºC to prevent changes in its composition.

To determine the colour, the fillets were exposed to the air for 45 minutes to allow oxygenation and the measurement was subsequently taken.

3. Physical-chemical determinations

The pH was measured 45 minutes and 24 hours post mortem by direct measurement with a Crison 2002 (Crison Instruments S.A., Barcelona, Spain) pH-meter with a penetrative electrode. The water activity was also determined by direct measurement with a Decagon Devices CX-2 (Decagon Devices Inc., Pullman, WA, USA) dew point water activity meter. The water retention capacity (loss of weight due to pressure) was determined by calculating the percentage of water that was liberated by the meat when it was subjected to a pre-determined amount of pressure in accordance with the method described by Grau and Hamm (1953) and modified by Sierra (1973). The humidity was determined in accordance with Regulation ISO R-1442 (1973). The fat was extracted with 40-60º petroleum ether in accordance with Regulation ISO R-1443 (1973). The total nitrogen quantity was determined utilizing the Kjeldahl method (AOAC 1980, method 16245). The nitrogen protein content was obtained multiplying the total nitrogen value by a factor of 6.25. The ash content was determined by the method described in Regulation ISO R-936 (1973).

4. Colour determination

The instrumental measurement of the colour was taken on a loin fillet employing a Konica Minolta (CM-2600d) mobile spectrophotometer. D65 brightener was used and measurements were taken under reflective conditions. The parameters were L* (Luminosity), a* (redness) and b* (yellowness).

5. Statistical treatment

To determine the physical-chemical composition and colour, the analysis was made on the variance (ANOVA) of a factor to study the influence of the configuration of the animal. When the interaction was significant, the averages were compared using the Student-Newman-Keuls test by means of the SPSS 17.0 for Windows XP (License UCLM 7876875) statistical program.

III – Results

The average values and standard deviations of the compositional physical-chemical parameters of the male pork loin obtained from the cross between the Duroc and Iberian pigs of various configurations are show in table 1 (A: light configuration; B: medium configuration; C: high configuration).

Table 1. The average values and standard deviation of the compositional physical-chemical parameters of the male pork loin obtained from the cross between the Duroc and Iberian pigs of various configurations

	A	**B**	**C**
Pork loin pH 45 minutes	6.09±0.27	6.28±0.21	6.17±0.23
Pork loin pH 24 hours	5.53±0.12	5.59±0.28	5.62±0.22
Ham pH 45 minutes	6.43±0.32	6.54±0.35	6.17±0.11
Pressure-induced weight loss (%DM)	10.79±2.13	9.40±2.23	12.20±4.04
Water activity	0.994±0.001	0.995±0.003	0.994±0.001
Moisture	69.30±1.62	68.94±1.36	67.52±3.57
Protein (%DM)	71.76^{a}±0.86	70.62^{a}±0.90	75.18^{b}±1.14
Fat (%DM)	27.79^{a}±5.17	20.81^{b}±4.23	19.64^{b}±4.67
Ash (%DM)	3.66±0.26	3.51±0.37	3.45±0.47

DM: Dry matter.
Superscripts (a,b) in any row and for the same factor denote significant differences (P<0.05) according to the Student-Newman-Keuls test.
A: light configuration; B: medium configuration; C: high configuration.

Significant differences were not found (P<0.05) for the pH values, pressure-induced weight loss, water activity, moisture or ash content; similar values were shown for the various configurations. However, significant differences were found (P<0.05) for protein and fat content. The pigs with light configuration (A) showed the highest fat content while the pigs with high configuration (C) showed the highest protein content. These results can be explained by the fact that at a higher configuration, there is a higher quantity of muscle and thus higher protein content and lower fat content is expected.

The average values and standard deviation for the parameters that define the male pork loin colour obtained from the cross between the Duroc and Iberian pigs of various configurations are shown in Table 2. Significant differences were found (P<0.05) for the value of a* (redness), being higher in the males of light and medium configuration.

Table 2. The average values and standard deviation for the parameters that define the male pork loin colour obtained from the cross between the Duroc and Iberian pigs of various configurations.

	A	B	C
L* (Lightness)	50.56±2.59	50.16±4.33	50.12±5.71
a* (redness)	5.35[a]±1.53	4.99[a]±0.92	3.69[b]±1.35
b* (yellowness)	13.75±1.32	14.04±3.52	13.39±2.05

Superscripts (a,b) in any row and for the same factor denote significant differences ($P<0.05$) according to the Student-Newman-Keuls test.
A: light configuration; B: medium configuration; C: high configuration.

IV– Conclusions

(i) Similar values were found for the pH, loss of weight due to pressure, water activity, humidity and ash content for the three lines of varying configurations that were studied.

(ii) The light configuration line (A) showed the highest fat content while the high configuration line (C) showed the highest protein content and lowest a* (redness) value.

Acknowledgments

The authors would like to thank the University of Castilla-La Mancha and the Agroibéricos de Raza S.L. company for financing the project and for supplying the specimens necessary to complete this study.

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Differences in productive parameters and carcass traits of three sires from Duroc x Iberian cross selected according to their conformation

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Abstract. The aim of this study was to determine the influence of animal selection on production parameters of the offspring from three Duroc (DU) boars selected according to their anatomical conformation. Thus, productive parameters and carcass traits were determined in 29 barrows from the offspring of DU x Iberian, descendants of three DU boars mated with Iberian sows. Sire A came from a DU boar with a low anatomical conformation, sire B from a DU boar with a middle conformation and sire C from a DU boar with a high conformation. All animals were reared on the same farm in intensive regime and fed with three types of feed at different stages of growth. Birth weight and average daily earnings at each stage of growth were determined. Furthermore, carcass, loin and ham weight, and back fat thickness were measured. The sire C showed a greater weight gain in the last phase of growth and total weight gain (weight at slaughter/days of life), this parameter was lower for sire A. Moreover, the carcass weight was higher for sire C and lower for sire A, which also differed by their lower weight of ham. The back fat thickness was higher for sire C, followed by sire B and finally sire A.

Keywords. Duroc x Iberian – Anatomical conformation – Productive parameters – Carcass.

Différences entre les paramètres de production et les caractères de carcasse chez trois lignées croisées Duroc x Ibérique sélectionnées en fonction de leur conformation

Résumé. *L'objectif de cette étude a été de déterminer l'influence de la sélection animale sur les paramètres productifs des premières lignées originaires de trois mâles de la race Duroc sélectionnés selon leur conformation anatomique. Par conséquent, les paramètres productifs et de carcasse ont été déterminés sur 29 mâles castrés provenant du croisement DU x Ibérique, descendants des trois mâles DU et de femelles Ibériques. La lignée A provenait d'un mâle DU de basse conformation anatomique, la lignée B d'un mâle DU de conformation moyenne et la C d'un mâle de conformation élevée. Tous les animaux ont été élevés dans la même ferme dans un régime intensif et nourris de trois types d'aliments selon la phase de croissance. Le poids des animaux à la naissance et les gains moyens quotidiens dans chaque phase de la croissance ont été déterminés. D'autre part, on a analysé le poids de la carcasse, de la longe et du jambon et l'épaisseur du gras dorsal. La lignée C a montré un plus grand gain de poids dans la dernière phase d'alimentation et le plus grand gain de poids total (poids à l'abattage/jours de vie), tandis que ce paramètre a été inférieur pour la lignée A. Enfin, le poids de la carcasse a été supérieur pour la lignée génétique C, et a été le plus faible chez la lignée A, qui s'est distinguée aussi pour avoir un poids plus petit de jambon. La quantité de gras dorsal a été supérieure pour la lignée C, suivie de la lignée B et de la lignée A.*

Mots-clés. *Duroc x Ibérique – Conformation anatomique – Paramètres productifs – Carcasse.*

I – Introduction

Because of consumer preferences and improved productive efficiency, the objective over the past few decades of genetic selection in pigs has been to increase the lean fat percentage of the carcass. This has caused a decrease in dorsal fat content and has led to a decrease in intramuscular fat content. Such changes have had positive organoleptics results (Gispert *et al*.,

1997) but carcasses with extremely high lean fat content can cause technical difficulties during processing and conservation. It would therefore be prudent to perceivably elevate the fat levels of the carcasses.

Iberian pig products, especially ham, are highly regarded in Spain. In recent years, however, the consumption of fresh meat has increased significantly as an alternative to white pig meat because the shelf life is longer, its appearance deteriorates more slowly and it provides improved organoleptics features (Estévez *et al*., 2003).

However, the Iberian pig has a slow growth rate as well as a low reproductive rate. Thus, in order to increase its productivity, it is frequently crossbred with the Duroc white pig at a level of 50% to 75% (B.O.E., 3/11/2007). This raises its reproductive capability, improves the growth rate and increases alimentary efficiency and lean fat content (Dobao *et al*., 1986) without a significant reduction in the quality of the meat, meat products (López-Bote, 1998) or the parts of the pig that are highly regarded and considered of higher quality than others (García-Macías *et al*., 1996).

The overall objective of this study was therefore to determine the influence of the genetic lines of the male Duroc with various anatomical configurations in male pigs from the first descendants by means of the productive parameters and characteristics of the carcass.

II – Materials and methods

1. Animal selection

This study was carried out using pigs from three lines of Iberian pigs of a cross between Duroc and Iberian breeds (50%-50%). Three males Duroc were acquired from a genetic selection company and were of various anatomical configurations: light (A), medium (B) and high (C). These males were crossbred with selected Iberian Retinto females.

In total, 33 mothers were used and two descendants were taken from each in order to minimize the influence of the mother and to highlight the influence the males had on the descendants. To complete this study, 10 castrated males were used from the first descendants of each of the lines.

The artificial insemination and rearing of the animals took place on a farm. The weight and number of pigs corresponding to each mother were controlled during lactation, rearing and feeding.

In order to track each pig individually and to guarantee its traceability from birth to slaughter, the animals were identified utilizing two tags that were placed on each ear.

2. Growth control during lactation and rearing

The pigs were weighed as soon as they were born and at the end of the lactation stage which lasted between 14 and 25 days. The average daily weight gain of the pigs (ADWG) (kg/day) was calculated during lactation by subtracting the weight at birth from the weight after lactation and then dividing by the number of lactation days.

Once the lactation finished, the pigs were given feed at intervals of anywhere between 74 and 92 days. This type of feed is referred to as both “starter” and feed 1. This feed prepared the pigs for the change in food as well as the alteration in the way they were fed during the fattening stage. The ADWG was calculated in this stage during their feeding.

3. Pig growth control during the fattening stage

During this stage, the pigs were given two types of feed. At the beginning of the stage, they were given a type of feed with high protein content, referred to as feed 2. At the end of the stage, they were given a type of feed that had higher energy content, referred to as feed 3. This type of feed was used so that the pigs would develop a higher quantity of fat.

The animals were fed with feed 2 for 110 and 119 days and with feed 3 for 94 and 120 days. The ADWG was calculated for each type of diet.

4. Control during slaughter

Finally, the pigs were weighed before being slaughtered (at approximately 150 kg and 10-11 months) and the total daily weight gain (kg/day) was calculated in the following manner: weight at the moment of slaughter divided by the days of life.

Various quality parameters of the carcass were determined in the abattoir:

- Weight of the carcass before and after being aired out (kg), employing a hanging scale

- Quality of the ham and loin following the carving of the carcass. It was calculated by dividing the weight of the piece by the weight of the carcass after being aired out.

- Dorsal fat thickness in cm measured at the 5th rib.

5. Statistical treatment

An analysis was made on the variance (ANOVA) of a factor to study the influence of the configuration of the animal. When the interaction was significant, the averages were compared using the Student-Newman-Keuls test by means of the SPSS 17.0 for Windows XP (License UCLM 7876875) statistical program.

III – Results

The average values and standard deviation of the productive parameters of the male pig obtained from the cross between the Duroc and Iberian pigs of varying configurations are shown in table 1 (A: light configuration; B: medium configuration; C: high configuration).

Table 1. The average values and standard deviation of the productive parameters of the male pork obtained from the cross between the Duroc and Iberian pigs of various configurations

	A	B	C
Birth weight (kg)	2.11±0.52	1.93±0.21	2.13±0.43
ADWG lactation (kg/day)	0.12±0.08	0.14±0.04	0.16±0.02
ADWG feed 1 (kg/day)	0.40±0.05	0.40±0.05	0.41±0.04
ADWG feed 2 (kg/day)	0.60±0.10	0.64±0.12	0.57±0.07
ADWG feed 3 (kg/day)	0.51^{a}±0.07	0.57^{a}±0.13	0.68^{b}±0.14
ADWG total (kg/day)	0.51^{a}±0.03	0.53^{a,b}±0.03	0.55^{b}±0.05
Carcass weight (kg)	119.2^{a}±9.27	126.6^{a,b}±8.25	130.5^{b}±13.83
Pork loin weight (kg)	2.98±0.34	2.80±0.44	2.94±0.47
Ham weight (kg)	14.08^{a}±0.89	15.58^{b}±0.90	15.74^{b}±1.58
Dorsal fat thickness (cm)	5.6^{a}±0.51	6.3^{b}±1.24	7.0^{c}±1.74

ADWG: Average daily weight gain; Superscripts (a,b) in any row and for the same factor denote significant differences (P<0.05) according to the Student-Newman-Keuls test; A: light configuration; B: medium configuration; C: high configuration

The genetic line influenced the ADWG in the final feeding stage (feed 3), the weight of the carcass and of the ham, total ADWG and the dorsal fat thickness. These differences were minimally significant ($P<0.05$) for these productive parameters. The male pigs from line C therefore showed the highest ADWG in both the final feeding stage as well as overall. The pigs from genetic line A showed the lowest carcass and ham weight while the dorsal fat thickness at the 5th rib increased significantly as the genetic configuration of the animals rose.

IV – Conclusions

(i) Similar values were found for the birth weight and the weight of the loin, the ADWG during lactation and for the rearing stages and first feeding stage for the three lines of varying configurations that were studied.

(ii) The light configuration line (A) showed the lowest ADWG in feed 3 and the lowest carcass weight overall, the lowest carcass and ham weight and the lowest dorsal fat thickness. The high configuration line (C), on the other hand, had the highest ADWG during the final feeding stage, the highest ham weight and the highest dorsal fat thickness at the 5th rib.

Acknowledgments

The authors would like to thank the University of Castilla-La Mancha and the Agroibéricos de Raza S.L. company for financing the project and for supplying the specimens necessary to complete this study. They would also like to thank the Agropecuaria Altozano farm and the Gypisa abattoir for allowing access to their facilities.

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CONBIAND Network: Following the genetic contributions of the Iberian pigs in the American Creole breeds using microsatellites

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Abstract. The CONBIAND network is a consortium integrated by researchers from 19 countries of America and Europe involved in biodiversity, sustainability and conservation biology. Within this network, a working group for pigs has been constituted in order to clarify the pig colonization history. Creole pigs were originated from animals brought to America by Spanish and Portuguese explorers more than 500 years ago. They are the result of genetic drift, artificial and natural selection and migration. The main goal of this project is to characterise some pig Creole breeds and to establish their genetic relationships with Spanish breeds. Several international breeds have been added in order to determine their influence on the Creoles. We have analyzed 9 Creole breeds with 24 microsatellites. Genetic distances and genetic structure were analysed in order to establish the genetic relationships between the Creoles and the Spanish pig breeds. The results derived from this project will have a great social impact, as may lead to the official registration of those populations, which are not yet officially recognized as breeds due to a lack of genetic supporting studies, but have an unquestionable productive rule in marginal areas and in a subsistence agriculture context.

Keywords. Genetic structure – Rural development – Latin American pig – Genetic distance.

Réseau CONBIAND: Recherche de traces génétiques de porcs d'Espagne chez les porcs créoles américains avec des microsatellites

Résumé. *Le réseau CONBIAND est un consortium de chercheurs de 19 pays d'Amérique et d'Europe dont les buts et objectifs sont axés sur la coopération pour le développement scientifique, social et économique de l'Amérique Latine. Au sein de ce réseau a été formé un groupe de travail spécial pour étudier la biodiversité, la durabilité et la conservation des races locales de porcs. Les porcs créoles issus d'animaux introduits en Amérique par les conquistadors espagnols et portugais il y a 500 ans, sont le résultat de la dérive génétique, de la sélection artificielle et naturelle et la migration. L'objectif de ce projet est la caractérisation de certaines races de porcs créoles et l'établissement de leurs relations génétiques avec des races espagnoles. Nous avons utilisé plusieurs races internationales en vue de déterminer leur influence sur les créoles. Nous avons analysé 9 races créoles avec 24 microsatellites. Les distances génétiques et la structure génétique ont été analysées afin d'établir les relations génétiques entre les créoles et les races porcines espagnoles. Les résultats de ce projet auront un grand impact social, car ils peuvent appuyer la reconnaissance officielle de ces populations, qui ont une importance incontestable pour la production dans les zones marginales dans un contexte d'agriculture de subsistance.*

Mots-clés. *Structure génétique – Développement rural – Porcs d'Amérique latine – Distance génétique.*

I – Introduction

The initial introduction of pigs in America occurred in the second Christopher Columbus trip, in 1493. This fact was described by Fray Bartolomé de las Casas in his “Historia de las Indias”. According this report, eight pigs were introduced in the Antilles and there they increased their population intensely in few years because the absence of illness and predators. Those animals that arrived to America with the Spanish and Portuguese explorers could be the origin of the pig populations existing today in the American continent.

Microsatellite markers have proved extremely useful for the analysis of population structure and relationships, and have been widely used for genetic characterization of several species and populations including European pig breeds (Laval *et al.* 2000; Martínez *et al.* 2000; San Cristobal *et al.* 2006; Vicente *et al.* 2008). Nevertheless, information on native breeds of Creole pigs is still scarce (Pérez *et al.* 2004; Canúl *et al.* 2005; Souza *et al.* 2010), even though they possess unique characteristics in terms of adaptation, hardiness, and quality of products. In this study we used microsatellite markers i) to evaluate the genetic relationships between the 9 American Creole breeds studied and ii) to assess with different statistical tools the integrity and degree of admixture of these native swine breeds.

II – Materials and methods

Individual blood or hair samples were collected from 1080 representative animals of the 25 populations under analysis. Specifically, the breeds studied and corresponding sample sizes are shown in Figure 1. DNA was extracted according to the method described in Walsh *et al*. (1991). A panel of 24 microsatellite markers (FAO 2004) was used: IGF1, S0002, S0005, S0026, S0068, S0090, S0101, S0155, S0178, S0215, S0225, S0226, S0227, S0228, S0355, S0386, SW24, SW72, SW240, SW632, SW857, SW911, SW936 and SW951. Microsatellite markers were amplified according the methodology used by Vicente *et al*., 2008. PCR products were separated by electrophoresis on a ABI 377XL instrument (Applied Biosystems, Foster City, CA) according to manufacturer recommendations and allele sizing was accomplished by using the internal size standard GeneScan-400HD ROX (Applied Biosystems, Warrington, UK). Results of electrophoresis were read directly and interpreted with GeneScan and Genotyper software (Applied Biosystems, Applera Europe B.V.), respectively. Genetic divergence among breeds was estimated through DA genetic distances (Nei *et al.* 1983), with the POPULATIONS software (Langella 1999). The STRUCTURE v.2.1 software (Pritchard *et al.* 2000) was used to investigate the genetic structure of the 25 populations, in order to identify population substructure and admixture. Runs of 500000 iterations after a burn-in period of 300000 iterations were performed for each K, to determine the most probable number of clusters, as inferred from the observed genotypic data. The DISTRUCT v.1.1 software (http://rosenberglab.bioinformatics.med.umich.edu/distruct.html) was used to obtain a graphical display of individual membership coefficients in each ancestral population.

III – Results and discussion

Most American Creole pigs grouped together forming a different cluster from that the formed by the current Spanish pig breeds (Figure 1). These results are consistent with those found by Revidatti *et al*., (2010), using the same panel of microsatellites used in this study, and with those obtained by Souza *et al*., 2010, using the Illumina SNP porcine 60k chip. Creole pigs in Bolivia and U.S. breeds grouped in the same cluster that Spanish pig breeds while the Creole pig in Venezuela is not part of any defined cluster.

A population clustering assessed using Bayesian inference revealed that most American Creole populations studied show a high degree of admixture and the Pelón Mexicano from Yucatán showed a clear substructure (Fig. 2). At the other extreme, Mulefoot Hog, Guinea Hog and Wild

Mexican from USA, Criollo Venezolano, Boliviano and Argentino (Dry) showed no evidence of genetic admixture and substructure.

Fig. 1. Dendrogram constructed from DA genetic distance (Nei *et al*. 1983) among 25 pig breeds from America and Europe. Numbers are the percentage bootstrap values from 1000 replications of resampled loci.

Populations: MF: Mulefoot Hog (USA); GH: Guinea Hog(USA); PM: Pelón Mexicano (México); WM: Wild Mexico (USA); S: Criollo Salvadoreño (El Salvador); EC: Criollo Ecuatoriano (Ecuador); CU: Criollo Cubano (Cuba); VE: Criollo Venezolano (Venezuela); BO: Criollo Boliviano (Bolivia); A(W): Caacolero (Wet) (Argentina); A(D): Caracolero (Argentina); RE: Retinto, Iberian Pig (Spain); LAM: Lampiño, Iberian Pig (Spain); EN: Entrepelado, Iberian Pig (Spain); TOR: Torbiscal, Iberian Pig (Spain); MJ: Manchado de Jabugo, Iberian Pig (Spain); CM: Chato Murciano (Spain); NC; Negro Canario (Spain); F: Negro de Ibiza y Formentera (Spain); NM: Negro Mallorquín (Spain); ET: Euskal Txerria (Spain); C: Celta (Spain); DU: Duroc (International); LW: Large White (Spain); L: Landrace (Spain).

Fig. 2. Population structure of 25 pig breeds using model-based STRUCTURE program (Pritchard *et al*. 2000). Each animal is represented by a single vertical line divided into K colours, where the coloured segment shows the individual's estimated proportion of membership in that cluster. Black lines separate the populations labelled above the figure. Graphical representation of individual genotype membership coefficients (q) when K=18, the optimum K value estimated.

IV – Conclusions

The results suggest that Creole pigs constitute a differentiated group from any of the ancestral breeds. Some Creole breeds show certain level of admixture whereas other are genetically defined. The results derived from this project will have a great social impact, as may lead to the official registration of those populations, which are not yet officially recognized as breeds due to a lack of genetic supporting studies, but have an important productive rule in marginal areas and in a subsistence agriculture context.

Acknowledgements

Authors wish to express thanks to the different breeders associations and research groups who kindly provided biological samples used in this study. The authors wish to acknowledge the financial support received by the Project RZ00-15 of the Programa Nacional de Recursos y Tecnologías Agroalimentarias of Ministerio de Ciencia y Tecnología of Spain and the International Cooperation of the Diputación de Huelva (Spain)

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Alentejo pig breed nipple (*Sus ibericus*) preliminar scientific notula (I)

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Abstract. The Author discuss the theme of this scientific notula, the results are preliminary, just to underline the danger of extinction and reproductive goodness of this genetic line on the characteristic numerical productivity (Pn) of the sow without nipple Alentejo and the Iberian autochthonous pigs breeds, both belonging to the Iberian Stocks.

Keywords. Boar Alentejo – Sow Alentejo – Nipples – Numerical Productivity.

La race porcine Alentejo à papilles (Sus ibericus) : note scientifique préliminaire (I)

Résumé. *L'auteur aborde le thème de cette note scientifique, dont les résultats sont préliminaires, juste pour souligner le danger d'extinction et l'intérêt de cette lignée génétique pour la reproduction en raison de la caractéristique productivité numérique (Pn) de la truie d'élevage sans papilles des races de porcs autochtones de l'Alentejo et Ibérique, appartenant toutes deux au Tronc Ibérique.*

Mots-clés. *Verrat Alentejo – Truie Alentejo – Papille – Productivité numérique.*

I – Introduction

The Agricultural School, Polytechnic Institute of Beja, began its administrative activities (July 15, 1985) and academic (Academic Year 1986/87) and from the Academic Year 1993/94, first proceeded to the layout of the facility the holding of Alentejo Pig Breed Herdade Agropecuária das Rascas in System Camping in Parks (100 mx 40 m) outdoors. Furthermore, we developed R & D, (Oliveira, 2000, 2008), studying some of the major productive and reproductive traits of Sows Alentejo Breeds founders of the holding, as can be seen in reports of work on graduation (Andrade, 1997 and Costa, 2001) and community support.

The Agricultural School, Polytechnic Institute of Beja, and the Herdade Agropecuárioa das Rascas (area 147.50 ha), the reserved area will be holding Alentejo Pigs Breeds corresponds roughly to 1.5 ha, divided into 4 (four) parks, 1 - Parturition / Create, 2. Mating, 3. Rearing and 4. Finishing / fattening, respectively. So we left early in the installation of a herd of 10 (ten) Reproductive's Breeding, 9 (nine) gilts and a boar, Breed Swine Alentejana without nipples.

Thus, from 2005 to date, we decided to analyze and compare the numerical productivity (Pn) of Alentejo sow (without nipples), when mated with the boar Alentejo nipples (experimental test 2006-2009). It is noted that in the literature, the average Pn breeding of Alentejo sow is 6.5 piglets per litter.

Currently, the Herdade Agropecuária das Rascas has a herd of this pig breeds (Figs. 1, 2 and 3), consisting of nine (9) sows without nipples, six (6) piglets castrated, eight (8) young sows, 1 (one) boar nipples, two (2) Varrasquetes (young boars) nipples and 17 (seventeen) piglets, a total of 43 (forty three) heads of animals.

Indeed, the goodness of the Iberian breed boar's nipple is proven since the fifteenth century (Alonso Herrera, 1513), as indicated Clement *et al*. (2006), whereas in Alentejo Pig Breed ... "Professor Miranda do Vale, mention of a red pig (Pork Alentejo nipples), since 1920, but says that there are very few in number, so it is a strain endangered ". Also according to Miranda do Vale (1949), quoting Dr. Janeiro tells us: "Currently there is a variety of race Alentejo redhead (nipples), with one representing very small and tend to reduce further, perhaps to disappear ...". For that reason since 2006 we decided to carry out this experimental work, although preliminary, because it is a line in danger of extinction and the data and the results of the actual records are scarce (in this particular case of a single exploration), as is evident this line is in danger of extinction according Sieuve Monteiro (1985) and FAO-Report (1992 and 2002 and more recently in the research work presented in 2006).

II – Objective

The purpose of holding the Escola Superior Agrária do Instituto Politécnico de Beja is the preservation of indigenous livestock species lines, particularly the autochthonous Alentejo Pig Breed for didactic-pedagogic, R & D and community support.

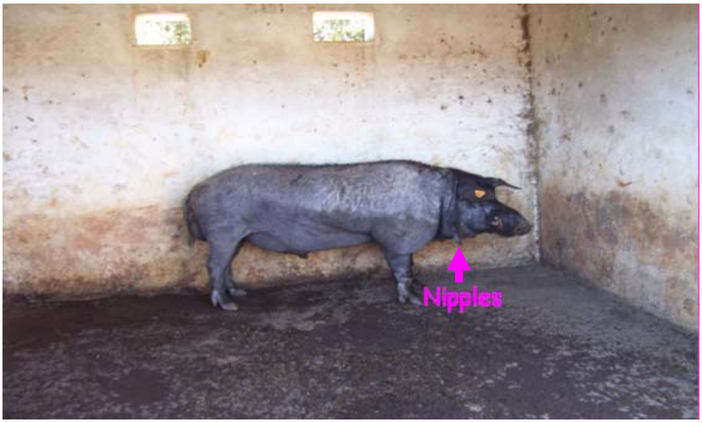

Fig. 1. Alentejo boar with nipples.

Fig. 2. Alentejo breed reproductives sows with nipples and without nipples.

Fig. 3. Alentejo piglets with nipples and without nipples.

III – Materials and methods

For this work, we use the breeding sows without nipples and with nipples above the farm where we note the numerical productivity (Pn) of their litters of sows, from 2006 until 2009, and proceeded to simple statistical analysis of variance and ANOVA with simple interpretation the results (Microsoft Office Excel, 2007)

IV – Results and discussion

Preliminary results from our test that we present in Table 1 and Fig. 4 (2006-2009), despite not differ significantly, there is a clear numerical superiority in Pn1 in relation to Pn, which we think is due to the effect of Alentejo Boar Breed nipples as referred Clement *et al*. (2006), concerning the goodness of reproduction (pleiotropic effect) of the Iberian Boar Breed nipples.

Table 1. Comparison of numerical productivity (Pn) of Alentejo reproductive sows (Alentejo boar breed without nipples vs Alentejo boar breed nipples) Agricultural School, Polytechnic Institute of Beja Exploração Agropecuária das Rascas

Year	Pn		Pn1		P	SIG
2006 – N = 21	N=18	4.72±1.02	N=3	7.33±0.58	0.541627	NS
2007 - N = 21	N=18	4.89±1.02	N=3	7.33±0.58	0.541381	NS
2008 - N = 20	N=15	5.13±0.99	N=5	7.40±0.55	0.541461	NS
2009 - N = 13	N=8	5.75±0.46	N=5	7.40±0.55	0.541223	NS

N - Number of breeding sows Alentejo without nipples and with nipples analyzed.
Pn - Numerical productivity of sows Alentejo breed covered by boar of Alentejo breed without nipples.
Pn1 - Numerical productivity of sows Alentejo breed covered by boar Alentejo breed nipples.
P - Probability. SIG - Significance. NS - Not significant.

V – Conclusion

Considering our results, although preliminary, there is the pleiotropic effect of the Boar Nipples on Productivity number (Pn1) of the breeding Sow Alentejo without nipples, so we recommend the implementation of more work of this nature in order to further study this parameter, taking into account their economic and financial importance, whatever the operating system of native pig breed Portuguese.

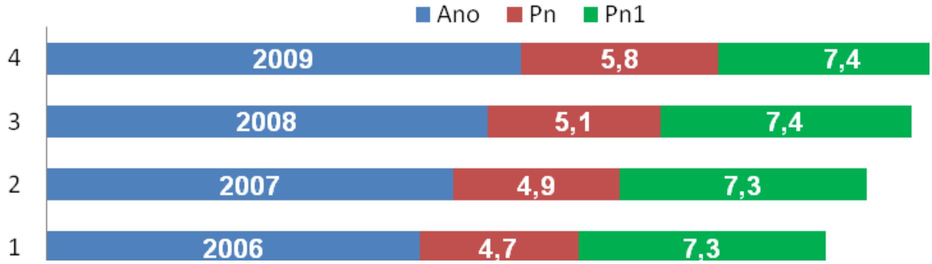

Fig. 4. Boar Alentejo breed nipples effect on numerical productivity (Pn1) of so Alentejo breed without nipples. Herdade Agropecuária das Rascas – Preliminar results.

Acknowledgments

The Author thanks all / those directly and indirectly contributed to this work

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Session 2
Production systems – Management

Production systems and sustainable management of pigs in the Mediterranean region

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Abstract. The Mediterranean pig production systems which enhances the value of endogenous resources and ecosystems are increasingly appreciated by the society, due to the beneficial role they play in the environment and the lives of animals (ethical quality), and to the associated nutritional and organoleptic quality. The type of breed (local Mediterranean genotypes), age and weight at slaughter, the climatic conditions, the use of regional raw materials (acorns, oaks, chestnuts, grass and agricultural by-products) and the exercise that the animals do during grazing, are the main differentiating factors of meat quality of Mediterranean pigs. In general the pigs destined for the Mediterranean high-quality products (PDO, PGI) are slaughtered at weights well above the age of sexual maturity (120-160 kg BW), when muscle growth has stabilized and at the expense of greater capacity of deposition and infiltration of intramuscular fat (+60% monounsaturated), an essential condition in order to develop the organoleptic characteristics highly valued in the market by the great appreciators. In these production systems, swine productivity and quality of final products depend on the interaction between genotype, environment, proper planning and implementation of animal husbandry on the farm: nutritional management, acclimatization and rearing conditions, optimization of reproductive techniques, health standards and levels of animal welfare, contribute to a greater extent to loss or gains of productivity and quality. Although they use production systems that are very close to natural methods, these holdings may incorporate some advanced technologies for management in a way to permit optimization of the quality and the economy of Mediterranean pig meat chain and also benefit the environmental balance of resources and landscape.

Keywords. Animal management – Production system – Performances – Quality.

Systèmes de production et conduite durable du porc dans la région méditerranéenne

Résumé. *Les systèmes méditerranéens de production porcine qui permettent d'améliorer la valeur des ressources endogènes et des écosystèmes sont de plus en plus appréciés par la société, en raison du rôle bénéfique qu'ils jouent pour l'environnement et la vie des animaux (la qualité éthique), ainsi que de la qualité nutritionnelle et organoleptique des produits. Le type de race (locales méditerranéennes), l'âge et le poids d'abattage, les conditions climatiques, l'utilisation de matières premières régionales (glands, châtaignes, herbe et sous-produits agricoles) et l'exercice que font les animaux au cours du pâturage, sont les principaux facteurs de différenciation de la qualité de la viande des porcs méditerranéens. En général, les porcs destinés aux produits de haute qualité (AOP, IGP) sont abattus à des poids bien supérieurs à l'âge de la maturité sexuelle (120-160 kg de poids corporel), lorsque la croissance musculaire est déjà stabilisée au détriment d'une plus grande capacité de dépôt et d'infiltration de la graisse intramusculaire (+60% de gras mono-insaturé), une condition essentielle pour le développement des caractéristiques organoleptiques très appréciées sur le marché par les grands connaisseurs. Dans ces systèmes de production méditerranéens, la productivité et la qualité des produits dépendent d'un grand nombre d'effets génétiques et du milieu et de l'interaction entre les deux, donc une bonne planification ainsi que le développement de bonnes techniques d'élevage sont essentiels aux différents niveaux de conduite des animaux: nutritionnelle, acclimatation d'élevage, reproduction, sanitaire, permettant de hauts niveaux de bien-être animal. Bien que ces systèmes de production soient proches des méthodes naturelles, les exploitations peuvent intégrer des technologies de pointe visant à l'optimisation de la qualité, l'économie des filières et l'équilibre des ressources naturelles et des paysages.*

Mots-clés. *Système de production – Techniques d'élevage – Performances – Qualité.*

I – Introduction

For many centuries in the Mediterranean region, the swine production systems and the husbandry of pigs were in perfect balance with agricultural practices and sustainable management of agro-forestry (oak groves *[montado* or *dehesa],* chestnut groves, forests and woodlands). The pig husbandry was performed according to the needs of animals, soil and climatic factors and sustainable use of agro-forestry. As a result of adaptation of animals to this system, the indigenous genotypes became less prolific and their carcasses were too fat – these genetic conditions allow the maintenance of reproductive capacity of sows under difficult environmental conditions, such as the harsh climate, the shortages or seasonality of food resources.

In the second half of the twentieth century, the increasing demand for lean meat and technological developments in pig husbandry combined with major social changes of that time, led to the intensification of pig production in Europe and, in parallel, to the abandonment of traditional production systems and the decline of local genetic resources. As a result serious problems arose related to environmental pollution, with animal husbandry and animal welfare, food safety has been questioned several times and it was necessary to develop further efforts in managing food risk to consumer health. The Mediterranean region has suffered particularly with these impacts, forestry and livestock grazing losing their importance, many farms were abandoned leading to processes of erosion and desertification of land, degradation of ecosystems, loss of biodiversity of fauna and flora, and as such the Mediterranean landscape has lost some of its typical features in some places.

However in the past decade traditional systems of pig production such as extensive or agro-silvo-pastoral (*Montanheira* or *Dehesa*) has shown a renewed alternative to intensive pig production, using less external inputs and increasing the modes of sustainable management of spaces agro-forestry. These production systems have demonstrated enormous advantages in terms of diversification of niche markets (high-quality local products sold at very high prices), contributing to the viability of a large number of companies and services, from production of pigs to trading of the products. To confirm this fact we find today, in any chain of hypermarkets or small gourmet food boutiques, hundreds of references of regional pork products (sausages, hams and sliced products) associated with indigenous genotypes and/or pig production systems that are typically Mediterranean (Portugal, Spain, Italy, France, Slovenia), many of which are protected under the European community legislation (DOP and IPG) – *see DOOR database for product names registered as PDO, PGI or TSG* http://ec.europa.eu/agriculture/quality/. In this system of pig productions – *Sustainable Mediterranean* – the logic of animal management is quite different from the logic of management applied in intensive pig rearing systems. The management must be as efficient as possible, particularly from the view point of the environment and animal needs and in the choice of technology best suited to the production levels and quality characteristics, otherwise, the cost of maintenance and amortization increases without any visible economic results as expected. In this paper we discuss some of the most important aspects in the management of Mediterranean pig with a view to a good managing product qualities and the environment.

II – Traditional pig production systems in the Mediterranean

These production systems – so-called sustainable – are developed in close relation with the biotic and abiotic factors, having as their primary specificity, the ability of the local breeds to adapt to different climates and rural ecosystems and the ability of these pigs to utilize the different agro-forest resources and turn them into high quality food products. Although they use production systems that are very close to natural methods, these holdings may incorporate some advanced technologies for management in a way to permit optimization of the quality and the economy of Mediterranean pigs and also benefit the environmental balance of resources and landscape. In what concerns the functioning and the utilization of food resources, the sustainable pig production systems in the Mediterranean area can be divided into two major groups.

(i) Extensive systems or agro-silvo-pastoral – fattening in *montanheira (PT)* or *dehesa arbolada (ES),* in which the indigenous pig, during the growth/finishing phases, utilize oak and acorns and less frequently the chestnuts, along with the grazing under the shaded areas. In general these systems predominate in the Mediterranean forests of cork oak and holm-oak (*Quercus suber* L.*, Quercus rotundifolia* L.), with the characteristic hot, dry climate and with medium to low soil fertility or with a low potential for agricultural production – manly in centre and south of the Iberian Peninsula, Corsica and Italy.

(ii) The complementary traditional farm or household production system, that does not make systematic use of forest food resources, because there are absent or scarce, but utilize more the products and by-products of local agriculture: cereals, tubers, vegetables, grass and a variety of seasonal fruits on a small scale. However, in cases of farms that have access to small woods or forests, the pigs can be placed here with a dual purpose to reap some vegetation and wild fruits (*Quercus* spp. *Castanea* spp.) and to contribute to the control of combustive material. These systems of poly-cultural production are usually associated with family farms of smaller dimensions situated in mountain regions, areas of high potential of plant biomass production in areas of micro-climates and wetter climates and soils of higher fertility – Northern Iberian Peninsula: Portugal (Trás-os-Montes, Minho), Spain (Galicia, Asturias, Basque Country), France and Italy.

The small size of the farms and the nature of the raw materials used in the traditional diet of these pig production systems has impeded the extension of the herd per farm, so in this sense, the management alternatives have been to take over the development of more intensive methods of production – intensive outdoor, camping or semi-confined – with the incorporation of technology and production factors external to the original system (e.g., cereals and food concentrates) adapted to the different physiological phases of the animals. For example, in northern Portugal small family farms associated with products from the breed with Bisaro of Origin (PDO and PGI) have increased in size by developing outdoor systems more or less intensive, often inserted in agricultural fields and small forests where the animals can graze on some herb, shrub and small fruits. In Fig. 1, we can observe the increase in average size of the Bísaro pig farms (breeding females registered in the studbook (LG), 2010), what has been achieved on the basis of increased outdoor settings.

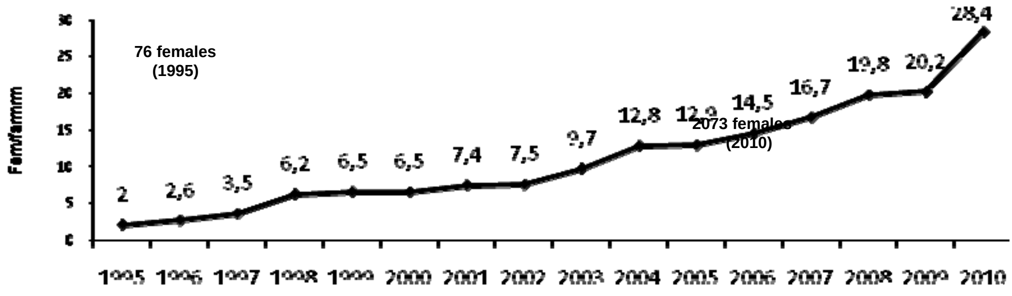

Fig. 1. Increase of the average size of the Bísaro pig farms (breeding females registered in the studbook (LG), data supplied by LG ANCSUB, 2010).

III – Animal management

The objective of animal management is to obtain a product with the highest quality and at the lowest cost, respecting the good production practices, animal welfare and the environment, taking into consideration the legal framework of the livestock sector and the market political context.

The animal husbandry is connected to how we do things to achieve a certain goal / objective, either from the standpoint of improving the effectiveness, efficiency and quality of processes and final products: guidance of fattening and finishing carcass; the conduct of reproduction and growth from birth to adult age and culling.

Animal management may have different levels of technological intervention, either using more or less land resources, more or less manpower or external production factors (capital). This sets the intensity of the use of natural resources, inputs and labor – the intensive regime, semi-intensive or extensive – which can be more or less from natural methods. The intensity of the regime used is related to the outputs ratios and obtained performances per unit of time or input, the level of technical and economic productivity and product quality. In general, compared to intensive production systems (livestock without land), traditional and sustainable pig farms in the Mediterranean involves large areas of land, using less technology and fewer external inputs.

Briefly, in an attempt to classify the main functions of animal husbandry, they may be divided on the type of technological activities and stages of the production cycle in which the animals are maintained: Global management – management planning in the farm; Planning – constitution of groups or lots of animals, social groups, number of lots, lot sizes, interval between lots; Acclimatization and buildings – housing conditions, environmental control, methods of animal containment; Culling herd – include genetic, annual replacement ratio, culling criteria, source of breeding, genotypes, genetic management – methods of crossbreeding and selection; Feeding management – *ad libitum*, restricted, semi-*ad libitum*, raw materials, diets, feeding and grazing methods, food distribution, water and watering; Reproductive management and breeding – breeding methods, assisted reproductive technology (induction, synchronization, super-ovulation, artificial insemination, pregnancy diagnosis, embryo transfer, cloning), farrowing, litters, weaning, yearling, age/weight first mating, breeding; Transport and slaughter management; Health management – preventive health, hygiene protocols, bio security measures and therapies; Management of pastures – rotations, fertilization, grazing, stocking rate; Ecosystem management – forests, mountains, "montado" or "dehesas" and specific ecosystems; Specific interventions in the animal – castration, cutting teeth and tails, markings, tattoos, chips, etc.

IV – Planning, animal management and quality improving

Animal management includes planning, programming and implementation of technological processes or tasks within the farms and has commonly been assigned a weight of over 25% in annual economic losses in the livestock sector.

A good animal management should provide a competitive advantage by incorporating technological factors and prove to be beneficial to the economy, market and society in general. In many cases, some animal management decisions seemingly simple can have very significant impacts on productivity and final quality, *eg.* the arrangement of lots, the organization of agricultural zones, the management of entry into "montanheira", the degree of surveillance of animals, assisted reproductive technologies, the simplicity of housing, bio security plans and others. In all cases they have to be carefully chosen and well executed, otherwise may cause damage and unexpected results.

Team work skills. The proper implementation of management techniques is the key factor of pig production and depends firstly the responsibility of team managers and players (business, engineers, veterinarians and technical staff and then planning capacity and resource management. In particular, depends on the technical and scientific knowledge held by staff, experience and training, organizational capacity and quality of implementation of each of the operations of management. Therefore, two similar farms, neighbouring each other, may have very different technical and economic results.

The pertinence of the use of certain technology management should be put up against the expected results, analyzing the cost/benefit and risk assessment. Very often, the results of operations of animal care are very different from that expected, because of poor technological applications, mismanagement or as a result of unforeseen incidents, or by direct influence of environmental factors or because of genetic interaction between animal and environment (G*A) (Hoste, 2003).

In some cases when a innovative technology is introduced on the farm, it is necessary to undertake a period of technological adaptation in the face of reality from the farm and the existing human resources, a topic that may justify the development of a research program applied for a trial period or a plan of adaptation and training of field staff.

Zoo-technical or biologic index. In short, the use of management methods requires a deep knowledge of the biology of animals and their responses to the different processes and technologies, considering the different environments and interactions. In pig production, these responses are monitored and evaluated using indicators in terms of performance efficiency, animal welfare, environmental impact, and product quality and food security, implying the existence of records and analyses tools - data collection field, computer records and analysis - that comprise the mathematical tools of management. A list of key indicators is presented in Table 1.

Table 1. Key indicators with economic interest mostly used in swine production.

Reproduction	Performances	Quality products
Semen quality, Age at puberty, Weaning-to-conception Interval, Farrowing interval, Piglets weaned, Embryonic and postnatal mortality, Numerical productivity, Ovulation rate, Litter size, Fertility rate.	Food intake, Average daily gain, Food conversion rate, Carcass losses, Live weight a certain age, Meat production per m^2, Milk production, Carcass income.	Carcass classification, Bacterial contamination, Fat (distribution), Intra muscular fat (I.M.F.), Oxidation potential, Lipid profile, Nutritional profile, pH 45 min; pH ultime, Water retention, Meat color, Tenderness, flavor and aroma.

These Indicators commonly used in swine production allow measuring the level of efficiency, effectiveness and quality of the work on the farm: ratios inputs/outputs; monitoring targets of productivity and quality and customers satisfaction. In terms of productive efficiency, the most commonly used parameters are the economic outcomes that relate the inputs consumption per unit of physical factors (per kg food, per sow, per m^2 of surface) or against periods of time (annual, monthly or per production cycle). Good examples of these indicators are the index of food conversion ratio (FCT), the average daily gain (ADG), the fertility rate (Tf) and numerical productivity (Pn). For the indicators of effectiveness and quality, parameters most used are related with the final product, such as the percentage of lean and fat and meat quality: water retention, nutritional meat profile, intra muscular fat content, colour, texture, tenderness and flavour.

Good management practices

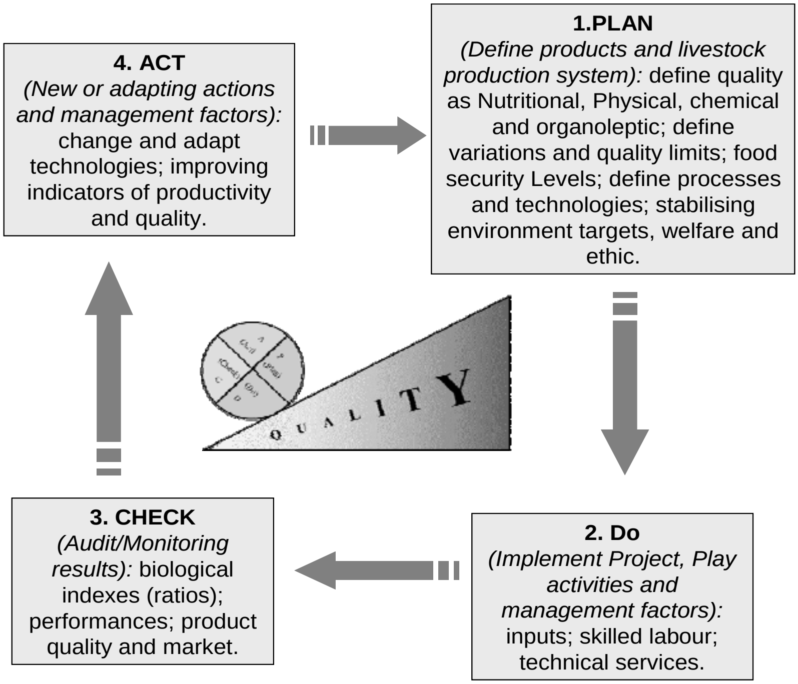

Fig. 2. PDCA cycle (Plan, Act, Check and Adapt) as a continual evaluation of the production system achieving efficient and permanent improvement of performances and quality.

The improvement processes and quality of pig production requires systematic monitoring and evaluation of results that will be obtained (via zoo-technical and quality indexes), aiming to find inefficiencies, loss of efficiency and reduced quality of products with the objective of upgrading and improving the methods of animal management used. The process of continuous quality improvement can be achieved by implementing a scheme of good quality management (Fig. 2) - the Deming's cycle or PDCA cycle (Plan, Act, Check and Adapt): 1. Plan (Define products and livestock production system; 2. Do (Project - Play activities and management factors); 3. Check (Audit/Monitoring results); 4. Act (New or adapting actions and management factors).

V – Animal husbandry, stress and animal welfare

Stress can be defined as a set of biological responses involving gene functions and hormonal responses and somatic when an individual receives a threat to its homeostasis. The threat is the factor of stress (cold, heat, equipment maladjusted to life, social interactions, pain, infection, hunger, fear) and can be rated according to their intensity and duration of its action (Moberg, 2000). Certain stressors (heat, social, fear) can alter animal behaviour and decrease food intake and/or uptake of nutrients in the gut, affecting body composition, immunity and animal welfare, and consequently affect the reproductive functions, growth and quality of final products. The negative consequences of stress are much higher when higher the intensity and duration of exposure of animals to stressors, and may have effects more or less serious, since the mere biological and regressive response, with loss of feed efficiency for repair animal welfare, to pre-pathological states or diseases, with loss of bodily functions, organ failure and death. Distress may occur at any stage of production process, during transport or in slaughterhouses. The main purpose of animal welfare management is to prevent and reverse the stress situations in order to improve the well-being, the performances and quality.

The implementation of most husbandry activities requires physical and psychological contact between man and animals likely to cause fear and pain, which in itself is a major stress factor. This is extremely important and raises important questions from the ethological and ethical point of view, related to the way we relate to animals, and the conduct that we have during the production process. Negligence, greed and accidents cause a large number of physical and metabolic lesions in the animals, which causes irreparable economic losses. However, in animal production, very often it is necessary to cause a short term pain (acute stress), to obtain the productive benefits in the long run. Some good examples of this are castration, health interventions, treatments and interventions for specific management, such as putting tags or tattoos. In these cases, whereas inevitable, good management practices indicate that these operations should be held as soon as possible, in safety to the operator and to animals, with minimal pain and done by trained personnel (Gregory, 1998). The way how animals are treated, have a great impact on the economy and competitiveness in the pork meat chains and are particularly sensitive in the public mind, consumer and society. That's why since 1991, the European Council published the first Directive setting out specific minimum standards for the welfare of pigs (Directive 91/630/EEC), which are legally binding on all member states. This legislation contains the basic principles of standards of protection and management of pigs for housing, handling and husbandry of the pigs: origin of stock, stockman ship and welfare, health and veterinary supervision, stock accommodation and handling facilities, feeding and water provision and farm cleanliness.

The outdoor pig rearing systems, the most used in the Mediterranean, are potentially more comfortable and have a lower initial investment cost, provide a greater enrichment of the environment, more space and opportunity to practice exercises of pigs. However, some disadvantages may also be considered: a greater risk of sunburn and heat stress, increased frequency of aggression and bullying. The outdoor hinders the feeding of sows individually, hinders the inspection of each animal and individualized treatment becomes more difficult. All these variables must be addressed and resolved through effective management and husbandry techniques should be more specific.

VI – Climatic factors in the Mediterranean and the husbandry of pigs

A major concern of pig production that should be considered in the Mediterranean region is the adaptation of the management techniques to the objectives of animal husbandry, the environment and the economics of the traditional and extensive or outdoor production systems.

The effect of heat stress causes a decrease in feed intake, alters the activity and social behaviour of animals, causes considerable losses of fertility, delayed growth, and changes the composition of carcass and meat quality. The problems of high temperatures during the transportation of animals to slaughterhouses increase the economic and quality losses of the final products (Gregory, 1998). For this reason the animal welfare during transport can not be neglected, and special attention should be done to the physical conditions of vehicles and animal handling, animal density, the distances travelled and time of travel. The very high temperatures, above the zone of thermal comfort of pigs, often associated with heat waves and periods of extreme drought are the main climatic stress factors affecting livestock pig production in southern Europe, estimating that their effects on gross annual loss of domestic product may exceed 20%: decrease of the performance, meat quality and reproduction losses, loss of production of cereals, pastures, fodder and forest fruits (Barbosa *et al.,* 2008).

The mobilization of body stores of lactating sows depends on the genotypes, the weather and the season and influences the prolificacy and fertility of sows, including anoestrus and the interval between weaning and ovulation (Santos Silva, 2006). The fact that native breeds have more body reserves and increased capacity to mobilize them evidenced a greater adaptive advantage of these animals to more extensive production systems, highly dependent on the variations of climate and food resources throughout the year. This ability to mobilize body reserves is linked to the adaptive capacity of local pig breeds to extreme environment conditions and can be understood by rusticity. Climate variations over the year and from year to year affect the results growth (ADG), deposition of fat and percentage of muscle and meat quality parameters as collagen and myofibrils fragment value (Santos Silva, *et al.,* 2006). Thus, under outdoor conditions, the seasonality is an important source of variation in performance and quality, the practices of animal husbandry should be based on the impact it can have on the agro-forestry resources and on the production of animals must adapt in terms of edaphologic and water resources, the potential of vegetal biomass production – cereals, grass and forest fruits (acorns, oaks and chestnuts) – with special attention to feeding management, grazing management and environmental adaptation of the animals to climatic variations.

VII – Animal husbandry and meat quality

The Mediterranean production systems which enhances the value of endogenous resources and ecosystems are increasingly appreciated by the society, due to the beneficial role they play in the environment and the lives of animals (ethical quality), and to the associated nutritional and organoleptic quality. The type of breed (local Mediterranean genotypes or crossbreeds), age and weight at slaughter, the climatic conditions, the use of regional raw materials (acorns, oaks, chestnuts, grass and agricultural by-products) and the exercise that the animals can do during grazing, are the main differentiating factors of meat quality of Mediterranean pigs. In general the pigs destined for the high-quality products (PDO, PGI) are slaughtered at weights well above the age of sexual maturity (120-160 kg BW), when muscle growth has stabilized and at the expense of greater capacity of deposition and infiltration of intramuscular fat (+ 60% monounsaturated), an essential condition in order to develop the organoleptic characteristics – aroma, flavor, color, brightness, texture – highly valued in the market by the great appreciators (Tirapicos Nunes, 2007).

VIII – Reproductive performances

For many centuries, swine production systems and the husbandry of pigs were performed according to the needs of animals, soil and climatic factors and sustainable use of agro-forestry. As a result of adaptation of animals to this system, the indigenous genotypes became less prolific and their carcasses were too fat – these genetic characteristics allow the maintenance of reproductive capacity of sows under difficult environmental conditions, such as the harsh climate, the shortages or seasonality of food resources. On the basis of genetic factors such as low genetic potential and the high consanguinity resulting from small herds that constitute the native breeds, are considered the main reasons for the low reproductive performances in local breeds. Reproductive traits such as the low ovulation rates and conception, high rate of embryo mortality and low survival rates after birth, failure in milk production and reduced postnatal growth are the main causes of loss in performance (Ollivier *et al.,* 2001). Other environmental factors related to seasonal climatic factors, with poor management or mismanagement of the production systems – poor nutrition, poor acclimatization, poor application of reproductive techniques, poor health standards and low levels of animal welfare – contribute to a greater extent to loss of productivity of the breeding sows. Limited by genetic factors, because selection of the reproductive traits are of low heritability, the improvement of environmental management and application of reproductive techniques are the most effective ways to improve reproductive outcomes in the short term.

Another option to improve reproductive performance in maternal lines in just one generation is the use of crosses between breeds (local x exotics breeds). In this case the breeds more used for obtaining breeding sows (F1) are the Large White breed and different lines of Landrace. However, these crossbreeding can alter the final quality of the products, especially if companies decided by supply pig meat chains only with purebreds. Duroc breed is most commonly used for terminal crosses with the aim of improving performances of growth and carcass (Martínez Serrano, 2008). In this case, the improvements are also obtained in terms of survival rate of piglets, as they have more vitality and viability due to the phenomenon of heterosis.

IX – General conclusions

The future of the Mediterranean production systems based on Multiple Use of Forest – pig meat production, management of combustible material, energy production, timber production, fruits, bark, biodiversity, agriculture, recreation and landscape – depend on the political and social consciousness of the local populations, the quality of the multifunctional management of agro-forestry resources and the maintenance of a healthy original ecosystem. As a general rule of good planning and animal management, the economic benefits of the pig production management practiced should be higher than the accrued costs of its introduction and should not interfere with the final quality of the products.

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The relative role of ovulation rate and embryo losses on prolificacy of Iberian sows

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Abstract. The influence of ovulation rate and embryo viability on the lower prolificacy of Iberian sows when compared to modern commercial breeds was firstly assessed in 78 non-pregnant cycling Retinto females showing a mean number of 12.7±0.2 corpora lutea (CLs). However, 66.1% of the females had ovulation rates under the mean (9.9±0.2 CLs); the remaining sows had 16.9±0.4 CLs (P<0.05). Such distribution was confirmed in 23 pregnant Retinto sows having 14.8±2.4 CLs; 69.5% of the females had lower ovulation rates (11.8±1.0 CLs), the remaining sows had 18.2±3.9 CLs (P<0.05). However, sows having high ovulatory rates showed, at Day 35 of pregnancy, a high incidence of CLs regression and embryo losses. Such effects were not found in females with low ovulation rates and, thus, number of viable embryos was finally similar in both groups (8.2±1.0 and 8.4±1.0). These results, indicating that prolificacy of Iberian sows is more influenced by embryo losses in the first third of pregnancy than by ovulation rate, was confirmed by studying 18 females of a prolific Retinto x Torbiscal strain. Mean ovulation rate was 21.3±0.5 CLs, but embryo losses reached a mean of 46.6%; the incidence of embryo mortality correlated linearly with ovulation rate (r=0.819, P<0.01).

Keywords. Embryo mortality – Ovulation – Pregnancy – Swine.

Le rôle relatif des taux d’ovulation et des pertes d’embryons sur la prolificité des truies Ibériques

Résumé. *L'influence des taux d'ovulation et de la viabilité des embryons sur une prolificité inférieure des truies Ibériques par rapport aux races commerciales modernes a été tout d'abord évaluée chez 78 femelles de Retinto non gestantes présentant un nombre moyen de 12,7 ± 0,2 corps jaunes (CJs). Toutefois, 66,1% des femelles avaient des taux d'ovulation sous la moyenne (9,9 ± 0,2 CJs); les autres truies avaient un taux de 16,9 ± 0,4 CJs (P <0,05). Cette distribution a été confirmée chez 23 truies Retinto gestantes ayant un taux de 14,8 ± 2,4 CJs ; 69,5% des femelles avaient des taux inférieurs d’ovulation (11,8 ± 1,0 CJs) et les truies restantes présentaient un taux de 18,2 ± 3,9 CJs (P<0,05). Toutefois, les truies qui avaient des taux d’ovulation élevés ont montré au jour 35 de la gestation une forte incidence de régression de CJs et de pertes d’embryons. Ces effets n'ont pas été rencontrés chez les femelles ayant des taux d'ovulation faibles et, par conséquent, le nombre d'embryons viables a été finalement similaire dans les deux groupes (8,2 ± 1,0 et 8,4 ± 1,0). Ces résultats, indiquant que la prolificité des truies Ibériques est plus influencée par les pertes d'embryons dans le premier tiers de la gestation que par le taux d'ovulation, ont été confirmés par l'étude de 18 femelles d'une race prolifique Retinto x Torbiscal. Le taux d'ovulation moyen était de 21,3 ± 0,5 CLS, mais les pertes d'embryons ont atteint une moyenne de 46,6%; l'incidence de la mortalité des embryons est en corrélation linéaire avec le taux d'ovulation (r = 0,819, P <0,01).*

Mots-clés. *Mortalité embryonnaire – Ovulation – Gestation – Porc.*

I – Introduction

The Iberian pig, like other Mediterranean swine breeds, is characterized by a clear predisposition to fat accumulation under its skin and among the muscular fibres (Nieto *et al.* 2002). This pattern of fat storage has been found not only in Iberian pigs but also in other animal species and even in humans, being named as *thrifty genotype* (Neel, 1962). The *thrifty genotype* is an adaptive mechanism to the environment, allowing accommodation to seasonal cycles of feasting and famine. The ability to store excess fat enables survival during periods of scarcity, but individuals with *thrifty genotype* become obese when food is in excess.

The abundance of fat in Iberian pigs causes an increased secretion of leptin when compared to lean swine breeds (Fernandez-Figares *et al.,* 2007), which has been also described for the Mangalica pig (Brüssow *et al.,* 2008). Leptin is the hormone produced in the adipose tissue for regulating appetite and food intake (Zhang *et al.,* 1994, Houseknecht *et al.,* 1998). The hormone was not identified until 1994 when cloning of the mouse obese gene was achieved (Zhang *et al.,* 1994). The obese mouse (ob/ob mouse), discovered in 1949 (Ingalls *et al.,* 1950), has deficiencies in leptin secretion and is characterized as being grossly overweight, due to high food consumption and scarce physical activity, and, additionally, was established as hyperglycemic, hyperlipidemic, and hyperinsulinemic. Consequently, these mice have been extensively used as a model for obesity.

However, in spite of high leptin secretion, it has been found that the Iberian pig has a gene polymorphism of the leptin receptors (LEPR) with effects on food intake, body weight and fat deposition (Ovilo *et al.,* 2005; Muñoz *et al.,* 2009); as a consequence, Iberian LEPR alleles increase insatiability and obesity. The same syndrome has been also described in human medicine and named as *leptin resistance*; some obese individuals have elevated leptin levels but fail in suppressing feeding (Martin *et al.,* 2008; Myers *et al.,* 2008) due to LEPR polymorphisms (Mizuta *et al.,* 2008).

Obesity and obesity-associated hyperleptinaemia by *leptin resistance* in humans have been linked to reproductive disorders (Metwally *et al.,* 2008; Brewer *et al.,* 2010). Leptin also has a key role in reproduction, acting through its receptors in hypothalamus, pituitary, ovary and endometrium (Cioffi *et al.,* 1997, Yu *et al.,* 1997, Gonzalez *et al.,* 2000, Duggal *et al.,* 2002, Watanobe 2002, Welt *et al.,* 2004). In fact, the obese syndrome also affects reproductive function (Lindström, 2007); a consequence of which both male and female ob/ob mice are infertile. Infertility is a direct consequence of leptin deficiency; thus, the administration of leptin restores weight, metabolic function and fertility (Barash *et al.,* 1996; Pallares *et al.,* 2010). Other strain of obese mice, in this case with monogenic deficiencies in LEPR ($Lepr^{db/db}$), is also infertile (Tartaglia *et al.*, 1995). In women, hyperleptinaemia has been related to menses irregularities, chronic oligo-anovulation and infertility; may be through a direct impairment of ovarian function (Pasquali and Gambineri 2006, Pasquali *et al.,* 2007), leading to alterations of granulosa cell function and follicle development (Fedorcsák *et al.,* 2000, Pasquali *et al.,* 2006), or may be through alterations in early-pregnancy and implantation (Bellver *et al.,* 2007).

The Iberian pig is also characterized by a lower reproductive efficiency, specifically a lower prolificacy (López-Bote, 1998), than modern commercial breeds; the same has been found in the Mangalica breed (Rátky *et al.,* 2005). Prolificacy in swine, as in other multiparous species, may depend on ovulation rate and/or embryo losses during pregnancy. Thus, the objective of this study was to characterize, for the Iberian breed, the consistency of ovulation rate and, thereafter, the incidence of embryo losses between Days 21 and 40 of pregnancy; a critical period that comprises from achievement of trophoblast attachment and implantation to completion of the transition from late embryo to early foetal stage (Ashworth *et al.,* 2006; Whittemore and Kyriazakis, 2006). For evaluating effects of the strain, pregnant females from purebred Retinto and a recombinant congenic strain with 75% Retinto and 25% Torbiscal (commonly used for breeding in practice) were compared.

II – Materials and methods

A total of 119 nulliparous Iberian sows, with a mean age of around 40 weeks and no previous evidence of health problems and adequate pathogen-monitoring reports, were used; all of them were genotyped for LEPR gene polymorphisms, as previously described (Ovilo *et al.,* 2005). Animals were housed indoors, in passively ventilated pens with concrete slatted floors, at either the Centro de Pruebas de Porcino (CPP, ITACyL, Hontalbilla, Segovia, Spain) or the INIA (Madrid, Spain). These facilities meet the local, national and European requirements for Scientific Procedure Establishments.

Assessment of ovulation rate was performed in a first experiment, by the observation of luteal structures in spontaneous non-induced oestrous cycles from 78 non-pregnant Retinto females reared at the CPP facilities from weaning. Ovaries were obtained at the slaughterhouse and, immediately after removal of the genital tracts, ovulation rate was determined by assessing the presence and number of luteal structures (*corpora haemorraghica*, *lutea* and *albicans*; Fig. 1).

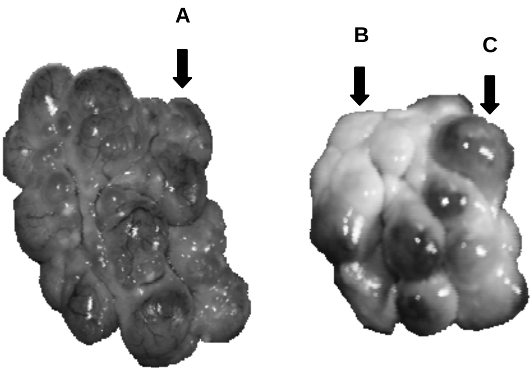

Fig. 1. Ex vivo image of corpora lutea (A), albicans (B) and haemorraghica (C).

Assessment of the relative roles of ovulation rate and embryo losses on prolificacy was performed in two consecutive experiments using 23 pregnant sows of Retinto strain and 18 pregnant Retinto x Torbiscal crossbreed females. These animals were treated, for cycle synchronization and breeding, with 20 mg of the progestagen altrenogest (Regumate®, Intervet International, Boxmeer, The Netherlands), daily for 18 consecutive days, by individually top-dressing over their morning feed; the treatment being initiated irrespective of the stage of the cycle. Oestrus detection was performed twice daily, from 24 h after progestagen removal; both inspection of the vulva for reddening and swelling (pro-oestrus) and control of the standing reflex (oestrus) in contact with a mature boar were performed. Sows were inseminated 12 and 24 hours after oestrus detection. Entire genital tracts were collected, between Days 21 (9 Retinto females) and 40 of pregnancy (14 Retinto and 18 Retinto x Torbiscal females), for evaluation of ovulation rate and characterization of conceptuses. Ovulatory sites in the ovaries were assessed for determining ovulation rate and evaluating morphologically normal and regressing corpora lutea. Thereafter, contents of the uterus were exposed and implantation sites and viable and non-viable conceptuses were recorded (Fig. 2) and compared to the number of corpora lutea.

For the statistical analyses, data from both pregnant and non-pregnant sow were summarized to characterize ovulation rate. Thereafter, data obtained were grouped according to genotype and day of gestation and the effects of these variables on number and characteristics of corpora lutea and conceptuses were tested by analysis of variance (ANOVA). Possible relationships between number and characteristics of corpora lutea and conceptuses were tested by Pearson correlation analysis and Spearman nonparametric correlation tests for non homogeneous variables. The parameter values were expressed as means ± SEM, and statistical significance was accepted for P< 0.05.

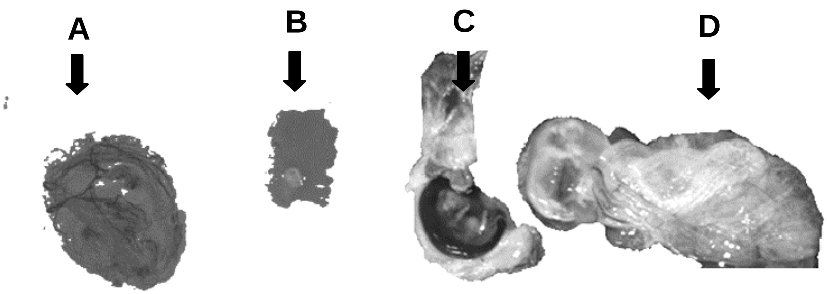

Fig. 2. *Ex vivo* image of viable (A and D) and non-viable (C and B) embryos at Days 21 (A and B) and 35 (C and D) of pregnancy.

III – Results

The assessment of the number of luteal structures in the non-pregnant cycling Retinto females showed a mean ovulation rate of 12.7±0.2. However, two different groups were found. Most of the females (66.1%) had ovulation rates under the mean (9.9±0.2), whilst the remaining sows (33.9%) had ovulation rates above the mean and significantly higher than in the first ones (16.9±0.4, P<0.05). Such distribution was confirmed when evaluating the ovulation rate in the 23 pregnant Retinto sows. The mean number of luteal structures was 14.8±2.4 CLs; 69.5% of the females had lower ovulation rates (11.8±1.0 CLs) and the remaining sows had 18.2±3.9 luteal structures (P<0.05). The ovulation rate in the Retinto x Torbiscal strain was higher than in both groups (pregnant and non pregnant) of Retinto females (21.3±1.4, P<0.005). The distribution of females with higher and lower ovulation rates was also different, since 50% of the animals had ovulation rates above and under the mean (25.4±1.4 *vs.* 17.1±1.1, respectively; P<0.01).

The incidence of embryo losses in the pregnant Retinto sows, and the comparison between 21 and 35 days of pregnancy, revealed that females with higher ovulatory rates showed a high incidence of luteal regression and embryo losses between Days 21 and 35 of pregnancy (around 50%) (Fig. 3). Thus, the number of viable embryos, at Day 35, was 8.2±1.0 in these females. On the other hand, such effects were not found in females with lower ovulation rates; in these sows the incidence of embryo losses was around 15% and the number of viable embryos at Day 35 was similar to the sows with higher ovulation rate (8.4±1.0).

In the Retinto x Torbiscal strain, embryo losses reached a mean of 46.6%, similarly to Retinto females with higher ovulation rate (Fig. 3). The incidence of embryo mortality correlated linearly with ovulation rate (r=0.819, P<0.01). Females with ovulation rate under the mean showed 31.2% of embryo losses; females with ovulation rate above the mean showed 55.9% of embryo losses. Thus, number of viable embryos in both groups were similar (11.8±0.7 *vs* 10.9±0.6), but higher than in Retinto females (P<0.01).

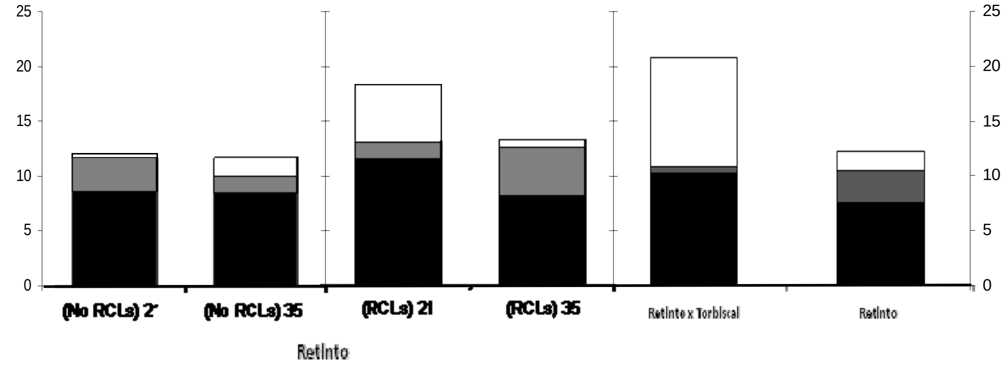

Fig. 3. Mean number of absent (white bar), degenerated (grey bar) and viable embryos (black bar) at Days 21 and 35 of pregnancy in Retinto sows with and without regressed corpora lutea (RCLs and NoRCLs) and comparison, at day 35-40 of pregnancy, between Retinto x Torbiscal and Retinto strains.

IV – Discussion

These results indicate that ovulation rate is not the main limiting factor for prolificacy of Iberian sows; prolificacy seems to be more influenced by embryo losses in the first third of pregnancy than by a lower ovulation rate.

When considering data about number of ovulations, the present results show a higher ovulation rate in the Retinto x Torbiscal than in the Retinto strain; which is in agreement with previous data obtained by farrowing observations (Suárez *et al.,* 2002a,b). Differences in the ovulation rate between the first and second replicates of Retinto may be related to the use of exogenous hormones in the second replicate. But, overall, our results suggest –besides a higher individual variability both in the Retinto purebred and in the Retinto x Torbiscal strain-two different "ovulatory behaviours". When evaluating the Retinto strain, most of the females (60%) had lower ovulation rates whilst some of them (40%) had high ovulation rates; the same was found in the Retinto x Torbiscal strain, although the distribution was around 50:50%. Females with higher ovulation rate also showed an intense luteal regression; thus, it seems that number of functional corpora lutea in pregnant Iberian sows was modulated either by a lower ovulation rate or by a lower quality of some of the corpora lutea in those females with higher ovulation rates.

Thereafter, prolificacy of pregnant sows was mainly hampered by differences in the number and viability of embryos. In swine, like in other mammals, prolificacy is determined by ovulation rate and/or embryo/foetal survival. The relative roles of ovulation rate and embryo survival has been extensively studied in the sow by using the Chinese Meishan pig, a very high prolific breed with average litter sizes 30 to 40% greater than European and American pigs (Bolet *et al.,* 1986; Haley and Lee 1993; Young 1993). These studies indicated, like in our study, that a higher number of corpora lutea in Meishan sows (Ashworth *et al.,* 1990; Haley and Lee, 1993, Anderson *et al.,* 1993; Christenson 1993), would not be enough to explain such higher prolificacy; issues related to a better embryo survival and developmental rate both at pre-implantational stages (Bazer *et al.,* 1988; Youngs *et al.,* 1993) and/or after implantation (Youngs *et al.,* 1993; Biensen *et al.,* 1999; Wilson *et al.,* 1999) would be definitive.

The present study, developed in Iberian sows, a different model with smaller litters than modern lean breeds, would be confirming the main role of embryo viability on swine prolificacy. Coincidentally, some authors have related deficiencies in reproductive outputs of obese females to alterations in early embryo development (Kawamura *et al.,* 2002; Fedorcsak and Storeng 2003), trophoblast function (Castellucci *et al.,* 2000) and endometrial receptivity (Alfer *et al.,* 2000; González *et al.,* 2000). The most recent studies in humans indicate an interrelationship between lower oocyte/embryo developmental competence and alterations in oviduct/uterine environment leading to deficiencies in early-pregnancy and implantation (Bellver *et al.,* 2007).

Implantation has been, and it is, widely studied in swine. Embryonic implantation is a crucial event in the establishment of pregnancy. Several factors have been reported to have roles in implantation and uterine receptivity (Giudice, 1994); within them, leptin. Leptin and LEPR (mRNA and protein) are expressed in the oviduct (Kawamura *et al.,* 2002; Craig *et al.,* 2005) and the endometrium (Gonzalez *et al.,* 2000; Kawamura *et al.,* 2002), suggesting possible involvement in endometrial receptivity for the developing embryo (Mitchell *et al.,* 2005). Moreover, the fact that LEPR are differentially regulated in implantation and inter-implantation sites suggests a regulatory role of the presence of an approaching embryo (Yoon *et al.,* 2005). Leptin and LEPR are known to be regulated by estradiol (Henson and Castracane, 2006) and embryonic estradiol is the signal for implantation in swine (Anderson *et al.,* 1993). Thus, it is possible to hypothesize that the different rate of protein secretion between implantation and inter-implantation sites described in the pig in the early 1990's would be regulated by estradiol and LEPR. We have to remind that Iberian pigs have leptin resistance and have a LEPR gene polymorphism disrupting processes of signal transduction; however, a possible relationship with implantation success may be tested by further studies.

V – Conclusions

These results suggest that prolificacy of Iberian sows is more influenced by embryo losses in the first third of pregnancy than by ovulation rate.

Acknowledgements

This work was supported by funds from INIA and ITACyL under the collaborative project CC07-014. The authors gratefully thank the CPP and INIA staffs for skilled technical assistance.

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The breeding of the main local pig breeds in Mediterranean Europe

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Abstract. The study reports the results of a survey on 7 local pig genetic types: Nero Siciliano in the Nebrodi territory of Sicily (Italy), Cinta Senese in Tuscany (Italy), Nero di Parma and Mora Romagnola in Emilia Romagna (Italy); "Nustrale" pig in Corsica (France); Greek pig (Greece). This study complements the analysis of a swine production characterized by strong territorial aspect as that of the improved pigs of Plana de Utiel-Requena, Valencia region (Spain). The paper reports statistics of technical and management aspects of the farms such as: farm size, land use, productive and reproductive parameters, marketing techniques. The comparison among breeds shows a substantial homogeneity of land use and a close link with the territory, although there are different levels of the rearing extensivity. In almost all cases the reproduction activity is the most critical element, as well as critical is the high variability of weights and slaughter age present in almost all areas under study. It is difficult to compare the several commercial strategies for selling products because they are characterized by very different levels of self-consuming. Niche production and high quality are the only elements common to all the surveyed farms.

Keywords. Local pig – Territorial survey – Farms features – Biodiversity.

Élevage des principales races de porc dans l'Europe méditerranéenne

Résumé. *L'étude indique les résultats d'une enquête portant sur 7 types génétiques de porc local : Nero Siciliano dans le territoire de Nebrodi en Sicile (Italie), Cinta Senese en Toscane (Italie), Nero di Parma et Mora Romagnola en Émilie-Romagne (Italie) ; "Nustrale" porc de Corse (France) ; Porc grec (Grèce). Cette étude complète l'analyse d'une production porcine caractérisée par une forte dimension territoriale comme celle du porc amélioré de Plana de Utiel-Requena, région de Valence (Espagne). Le document montre des statistiques sur les aspects techniques et de gestion des exploitations agricoles, tels que: taille des exploitations, utilisation des terres, paramètres de production et de reproduction, techniques de marketing. La comparaison entre les races montre une homogénéité substantielle de l'utilisation des terres et un lien avec le territoire, bien qu'il existe différents niveaux d'extensification. Dans presque tous les systèmes, la reproduction est l'élément le plus critique, ainsi que la grande variabilité de poids et l'âge d'abattage présents dans presque toutes les réalités étudiées. Il est difficile de comparer plusieurs stratégies commerciales pour la vente des produits parce qu'ils sont caractérisés par plusieurs niveaux d'autoconsommation. La production de niche et de haute qualité sont les seuls éléments communs à toutes les exploitations étudiées.*

Mots-clés. *Porc local – Enquête territoriale – Caractéristiques des exploitations – Biodiversité.*

I – Introduction

The Qubic project, funded by the EU with the MED program has the aim to verify the breeding of 7 genetic types of local pigs, in different areas of the European Union belonging to Mediterranean Regions. The partners involved in this project come from different countries: 3 Italians (Regione Sicilia, ARSIA-Regione Toscana, SSICA- Regione Emilia-Romagna), a Spanish partner (AINIA-Valencia), a Greek partner (INA-certh-Thessaloniki) and two French partners (INRA-Corsica and Chambre du Drome).

II – Materials and methods

1. Animals

Six types of local pigs have been examined. Four reared with extensive techniques:

1) *Nero Siciliano* in the Monti Nebrodi area (Sicily, Italy)
2) *Suino* "*Nustrale*" in Corsica (France)
3) *Cinta Senese* in Tuscany (Italy)
4) *Greek Pig* (Greece)

Two reared with semiextensive techniques:

5) *Mora Romagnola* in Romagna (mainly in province of Ravenna) (Italy)
6) *Nero di Parma* in province of Parma (Italy)

It was also carried out an analysis (with the aim of comparison) on pigs reared under intensive techniques for high quality production with strong territorial value:

7) Improved pig (Large-White, Landrace, Pietrain, White Belgian) in *Plana de Utiel-Requena* (Valencian Region, Spain)

Nero Siciliano

The "*Nero siciliano*" is mainly present in eastern Sicily, in the province of Messina, on Nebrodi mountain, so this breed is also called "Suino nero dei Nebrodi" (Chiofalo *et al*., 2007). The *Nero* pig has black skin, where strong black bristles, that can reach about 10 cm long, are implanted on neck, withers, back, loins. The rearing system of *Nero Siciliano* is mostly outdoors fed barley and field beans but a small number of subjects is also held in free range system fed natural vegetation of the undergrowth. There are 111 farms that rear this breed with a total of 644 sows and the Qubic survey was conducted on a sample of 36 farms.

Nustrale

The "*Nustrale"* pig breed is the local pig breed of the Corsican Island and it is present on the whole island (Casabianca *et al*., 2000). The regional breeders association (created in 1998) works very close to the association constituted (in 2001) to apply for a PDO for high quality cured meat products. The rearing system is extensive and based upon the use of local resources like pasturelands, chestnuts and acorns. The products are processed in a traditional way and often sold by the farmer himself. The breed was officially recognized in 2006 by the national authorities, which allows mentioning it as mandatory in the PDO specification.

The survey concerned 23 farms located in Corsica.

Cinta Senese

The "*Cinta Senese*" pig breed is native of Tuscany and it is farmed in the Region since immemorial time (AA.VV., 2004) This breed has black skin with a white belt on withers and front legs. Currently, its rearing is expanding both in the Region and outside; during the 80s, the breed has been threatened of extinction because of the strong competition of improved genotypes. The rearing of this breed is always outdoors on pastures and/or forest land with different levels of extensivation and animal load. The farming system varies from full free range to a more rational system that includes the use of shelters during the reproduction period. Official reports show the presence of 147 farms in Tuscany with a total of 1220 sows. The Qubic survey concerned a sample of 36 farms with approximately 681 sows, which represent more than one half of the total number of the reared sows.

Greek Pig

The Greek pig is a domestic breed, that looks like the wild boar and it survives in the northern areas of Greece. The rearing of this breed is at present in expansion whereas, during the 80s, the breed has been threatened of extinction because of the strong competition of improved genotypes and the crossbreeding with the wild boar. The dominant colour of this breed is black, but animals can be black-brown, brown with white grooves as well as black with white spots. They live generally on free range, grazing on oak-forests. Currently the number of sows is 350 units, distributed in 10 farms. The survey covered all farms located in Thessaly, C. Macedonia and Western Thrace.

Mora Romagnola

"*Mora Romagnola*" was widespread in the province of Forli and Ravenna, but also throughout the Romagna (Tassone and Fortina, 2003). The name "Mora" was codified in 1942 and it is due to its colour, dark brown tending to black. The salient morphological feature of the breed is the coat with pigmented skin (black or dark grey) on the back and in external areas of the limbs, while abdomen and inner faces of forearm and thigh are rosy. It presents black and tan coat with long bristles with divided and reddish tip. Today there are 46 farms registered in the herdbook with approximately 450 animals (300 females and 150 males). The survey covered 23 farms almost located in province of Ravenna.

Nero di Parma

The "*Nero di Parma"* pig is the result of a recovery project which began in the early 90's. The work of selection and study of morphological aspects resulted in the reconstruction of the genetic hybrid "Nero di Parma" (Sabbioni *et al*., 2009). From the 90's some breeders join together in a Consortium and developed, together with institutional bodies, a trademark to identify products derived from meat of "*Nero di Parma"* pig. The rearing of this breed is semiextensive and at present there are 841 females and 45 males registered. The survey concerned 20 farms in the Parma Province.

Improved pig of *La Plana de Utiel-Requena*

In this Spanish region take place an industrial type of pig breeding with a strong specialization of the various productive sectors so that in 114 farms, only 92 of them carry on fattening stage and 12 are multipliers or breeders. The employed breeds are: Large White, Landrace, Pietrain, Belgian White and the goal is to produce a light weight pig (about 110-120 kg), for fresh consumption and for the production of typical products of Requena. This production involves the use of natural products like lemon, orange, cinnamon, etc. In this situation the farm size, measured as number of sows, refers to classes that are completely different from those used for the local pig system because of only 20% of the surveyed farms raises less than 50 sows (representing only 1% of the total sows) and another 20% (57% of total sows) is in the class between 1500-2500 sows.

2. Monitoring

The project involved a detailed monitoring of the activity of the farms, of the production environments in which they operate, of the transformation phase and the enhancement of products and issues concerning the marketing of various productions. The survey was conducted on a sample of representative farms allowing to analyze the situation of reared breeds identified in a well-defined territory, which usually corresponds to the native area and / or maximum spreading area.

III – Results and discussion

The analysis of the parameters measured during the visits conducted in the various farms, allows some reflections to highlight the common or distinctive aspects. While postponing the consideration of individual reports for a more precise and detailed analysis of different situations. These are the main results obtained:

1. Farm size

The summary of data on Table 1 shows a remarkable homogeneity among the realities analyzed with regard to farm size, measured by the average number of sows. They are always realities based on a limited number of breeding pigs, with 30-40% of those managing less than 5 sows. One exception is the reality of the "Nustrale" pig, having a greater presence of medium-sized farms (6-50 sows). It is noteworthy to point out, in the case of the Cinta Senese in Tuscany (Bonanzinga et al. 2007) and Mora Romagnola in the province of Ravenna, the presence of large farms (at least for the local pig breeds), which indicates a tendency to move towards economies of scale. Also the reality of the Greek Pig production appears oriented towards medium-sized herds, with almost half of the farms with more than 100 sows.

As a comparison with the actual production of local pig breeds, it can be reported what emerges from the report on the Plana de Utiel-Requena.

Table 1. Distribution (%) of herds in dimensional classes on the basis of number of sows. In parenthesis the incidence of sows on the total

	Herd size (no. of sows)			
	1-5	**6-15**	**16-50**	**> 50**
Extensive rearing				
Nero Siciliano	37	33	30	
Cinta Senese	29(6)	38(17)	21(23)	12(54)
Nustrale	4 (1)	52(33)	44(66)	
Greek pig	10	10	30	50
Semiextensive rearing				
Nero di Parma	42(13)	42(45)	16(42)	
Mora Romagnola	37(7)	48(40)	10(23)	5(30)

2. Land use

The surveyed herds can be attributed to three types of rearing techniques: extensive, intensive and semi-extensive. The extensive type is the most representative and it is realized for *Nero dei Nebrodi, Nustrale, Cinta Senese* and *Greek Pig* breeds. In this case, there are medium- large herds, that often includes areas used for grazing pigs. The widespread presence of oak and chestnut, also allows the use of grazing for food. Sometimes the continuous presence of pigs within the wooded areas caused erosion and damages to plant roots. In these cases it is desirable to have a more rational management of these areas, pointing to the rotation of grazing areas and reducing the animal loading by excluding all the breeding animals and in the most critical cases also the lean pigs from grazing in the wood. As regards the semiextensive rearing, the concerned breeds are *Nero di Parma* and *Mora Romagnola*.

The farms have relatively small areas, the surface for grazing is present in a very small proportion and there are some open spaces for pig breeding.

3. Reproductive activity

Regarding reproductive activity, in most cases the number of weaned piglets was around 5-7 and the weaning age was around 60 days for the extensive farms. The considered breeds are rustic breeds and therefore their reproductive performance reflect the characteristics of these animals, usually not improved and therefore less productive.

Data obtained from breeds reared with semi-extensive techniques do not differ much from those of free range, while the improved pig from the Plana de Utiel-Requena highlight reproductive performance in line with the European standards for industrial pig breeds (2.42 birth / sow / year with an average of 20.5 piglets weaned) (Table 2).

Table 2. Reproductive performance

	Litter size			
	Born alive	**At weaning**	**Births/sow/ year**	**Weaned/sow/ year**
Extensive rearing				
Nero Siciliano	8.97	7.54		
Cinta Senese	6.82	6.16	1.59	9.80
Nustrale	6.87			
Greek pig	8.10	5.30	2.00	10.60
Semiextensive rearing				
Nero di Parma	7.40	5.41	1.35	7.30
Mora Romagnola	8.31	6.52	1.75	11.4
Intensive rearing				
Pig of Plana de Utiel-Requena	10.90	8.45	2.42	20.50

4. Productive parameters

The production of local breeds shows a wide variability of weights and slaughter ages. This variability is present within each farm, between farms within the same breed (because of production decisions and management) and between local breeds in relation to the intrinsic characteristics of growth, maturation and body size. It's very interesting the comparison of local breeds with the pig farming of Plana de Utiel-Requena which has a well-defined production target (light weight pigs), using improved pigs (standard rearing conditions) with the aim of meet the economic logic of the market that requires growth rates well calibrated and standardized.

Therefore, in this case age and slaughter weight do not fluctuate too much and the most frequent production is focussed on a pig weighing in the range of only 5 kg.

Regarding breeds reared in extensive conditions, it's possible to make some remarks and to establish a differential framework, as shown in the summary presented in Table 3.

The *Cinta Senese* breed is the one that achieves the highest slaughter weight (150 kg) which allows a good production of hams (Bonanzinga *et al*. 2007). *Nero dei Nebrodi* and *Nustrale* show an almost identical situation, reaching an average slaughter weight of 110-115 kg that guide the production mainly to various types of salami. Finally, the *Greek pig*, slaughtered at a very light weight (70 kg), is intended for fresh consumption, because in Greece there isn't a real tradition of processed pig meat products.

Regarding the breeds reared in semi-extensive conditions, the slaughter weight is significantly higher than that for animals reared in extensive conditions and varies from 160 kg for *Mora Romagnola* to 190 kg for *Nero di Parma.*

With these characteristics of carcasses it's possible to make all the types of productions following processing techniques typical of the native area especially ham, coppa, pancetta, etc.

Finally, in La Plana de Utiel-Requena pig production is addressed to lightweight pig (approximately 115 kg) at an average age of slaughtering of 6.5 months. The production is directed to the processing industry while a small percentage is intended for fresh consumption.

Table 3. Age and weight of slaughter

	Age (months)	Slaughter weight (Kg)
Extensive rearing		
Nero Siciliano	18	115
Nustrale	18	110
Cinta Senese	18	150
Greek pig	10	70
Semiextensive rearing		
Nero di Parma	17	190
Mora Romagnola	16	160
Intensive rearing		
Pig of Plana de Utiel-Requena	6.5	110

5. Destination of the products

The situation of commercial products of the 6 local pig breeds is extremely variable and it is hard to give an overview of the various trade flows (Table 4).

Table 4. Destination of the products

	Self Use	Direct sale/ restaurant	Butchers/ traders	Industry
Extensive rearing				
Nero Siciliano	3	97		
Cinta Senese	5	43	22	30
Nustrale	1	90	9	
Greek pig		58	32	10
Semiextensive rearing				
Nero di Parma		37	58	5
Mora Romagnola		23	20	57
Intensive rearing				
Pig from Plana de Utiel-Requena			18	82

Even if all of them are niche production, the share of auto-consumption is very low, reaching the peak in *Cinta Senese* (5%). Direct selling is possibly the most widespread method of marketing, particularly in the realities of *Greek Pig* (58%), of *Nustrale* pig, where it reaches more than 90%, and of *Nebrodi Black pig*, where it is the only commercial channel. For the other breeds also, however, the direct selling represents a good slice of the market (43% in the *Cinta Senese*, 37% in *Nero di Parma*, 23% in *Mora Romagnola)* and it often occurs with direct sales to private individuals or to agro-touristic farm going side by side together types of commerce such as restaurants, butcheries and shops. For the commercial realities of these last three breeds, however, it is also an ongoing a relationship with the processing industry that is being developed especially for *Mora Romagnola* and *Cinta Senese*. This last breed begun to direct the product towards few large Companies and GDO which are capable of enhancing the product, ensuring wide distribution anyway. The light pigs produced in the *Plana de Utiel-Requena*, however, has a commercial destination far less complex following the normal channels of industrial production which are the processing industry and butchers.

IV – Conclusions

The Qubic European project allowed to compare, for the first time, all the genetic types of native pig breeds of the Mediterranean Basin Area, except for the Iberian swine that for bureaucratic reasons has not been included in the Qubic project yet.

In recent years all indigenous pig breeds have been involved in programs and projects for recovery and valorisation using the European, national and regional funds, except for the Greek pig that has been identified with this project.

Almost all the concerned herds show an organization that provides the complete production cycle. While in the case of the *Mora Romagnola* pig breed surveys has been made in some farms (17%) that provide only the fattening stage. This last type is rare and rather temporary for the local breeds, because there is no need to specialize the farming of these breeds.

Farms surveyed are very similar even if the morphological characteristics were different, highlighting also significant differences in relation to their slaughter weight (from 110 of *Nustrale* to 150 kg of *Cinta Senese* up to 190 kg of *Nero di Parma*).

The slaughter weight is reached at different times and it shows a wide variability even within the same breed.

So there are margins to improve farming techniques without changing the characteristics of a typically extensive farming.

Finally, the relationship with the territory and in particular with the use of forests. In some situations the grazing in the wood is not much used, while in many others situations we can observe a strong animal loading on the woodland. Often the animals are placed permanently in the wood causing damages without gaining an advantage in terms of feeding.

This is certainly a problem and it could also become a limiting factor for the development of this type of farming.

Trying to make compatible the native pig breeding with a sustainable grazing in the wood is certainly the challenge for the next years, a challenge that will ensure the development of these important breeds and will guarantee the consumers about the availability of quality products that, because of their typical characteristics, will be very important to represent a given territory.

Acknowledgements

The research was supported by MED Territorial Cooperation Programme, funded by the European Regional Development.

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The link between breed, territory and product quality: the case of the Cinta Senese

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Abstract. The study reports the results of a survey on farming issues from a representative sample of Cinta Senese farms located in Tuscany. The paper reports statistics relating to technical and managerial aspects of farms such as: farm size, land use, feeding techniques, production and reproductive parameters, health management, marketing techniques. The information generated were then analyzed to identify the strengths and weakness of the system, while giving the real opportunities for development of the breed. Again, thanks to results of the survey, this paper describes the main types of salami made from Cinta Senese, indicating, also in this case, the strengths and weakness of the system, especially for what concerns the technical processing and marketing strategy. Particular emphasis is given to issues related to the management of the Protected Denomination Origin (PDO) that, for the Cinta Senese,was obtained, as a transitional, exclusively for the fresh meat.

Keywords. Cinta Senese – PDO – Territorial survey – Salami.

Le lien entre la race, le territoire et la qualité des produits: le cas de la Cinta Senese

Résumé. *L'étude montre, tout d'abord, les résultats d'une enquête sur les modes d'élevage d'un échantillon représentatif des exploitations de Cinta Senese dans le territoire de la région de Toscane. Le travail rapporte les statistiques concernant les principaux aspects techniques et de gestion de l'exploitation parmi lesquels: dimension de l'exploitation, usage du sol, techniques d'alimentation, modèles productifs et reproductifs, gestion sanitaire, techniques de commercialisation. Les résultats obtenus ont été analysés pour déterminer les points de force et de faiblesse du système en indiquant en même temps les opportunités réelles de développement de la race. Le travail décrit aussi les principales typologies de charcuteries produites avec Cinta Senese, en indiquant aussi dans ce cas les points de force et de faiblesse du système surtout en ce qui concerne les techniques de transformation et de commercialisation. Une attention particulière est accordée aux questions relatives à la gestion de l'Appellation d'Origine Protégée (AOP) pour la Cinta Senese, qui a été obtenue provisoirement exclusivement pour la viande fraîche.*

Mots-clés. *Cinta Senese – AOP – Enquête territoriale – Charcuterie.*

I – Introduction

The Cinta Senese, pig breed native of Tuscany, is farmed in the region since immemorial time; currently, its rearing is expanding also in outside regions after that during the years 60s-80s, it was in danger of extinction because of the strong competition of the improved genotypes. To date, the official statistics report the presence, in Tuscany, of 147 farms with 1,200 sows in breeding. The farming system varies from the completely free range, in which the animals are not supported by structures and receive minimal food supplementation, to the more rational system that uses shelters of varying complexity, especially for the reproduction phase, and food support, not far from the industrial systems. However, the rearing is always outdoors on agricultural land and/or forest with different levels of extensivation and animal loading. The wood used is the typical Mediterranean forest constituted primarily of deciduous oak (*Quercus cerris, Q. suber, Q. pubescens*) and evergreen (*Q. ilex*) and of chestnut trees. The success of the breed is being realized thanks to the special quality of its fresh meat and cured products. If properly addressed with adequate feeding, which has a strong influence on the fatty component, the organoleptic characteristics of its products are especially appreciated because of they are well distinguishable from those of the products originated by standard pig.

II – Materials and methods

The report relates on the rearing system of Cinta Senese and involves a representative sample of farms (35 on 147) (Fig. 1). Referring to the number of breeding animals, the sample includes about half of the sows reared in Tuscany (681 on 1200). This study wanted to notice technical information on breeding management, productive performances, marketing strategies, processing techniques and to point out breeders' technical and commercial needs.

The survey involved all the provinces of Tuscany except Massa Carrara, which has no farms of Cinta Senese. The distribution Cinta Senese farms by province is represented in Fig. 2: the majority of the farms is located in the province of Siena, while the lowest number of farms surveyed is located in the provinces of Lucca and Prato with just 2.7%.

Data were analyzed using SAS statistical software (SAS Institute Inc., 2003).

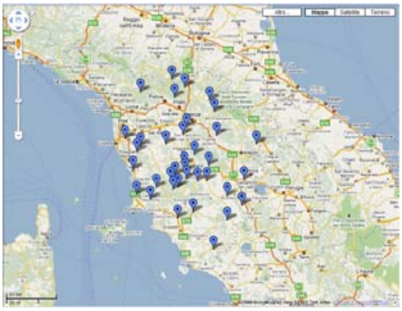

Fig. 1. Localization of farms included in the survey.

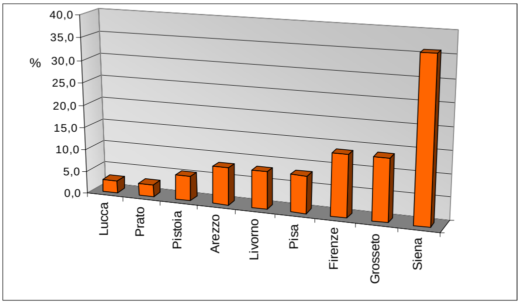

Fig. 2. Percentage of Cinta Senese farms by province.

II – Results

The most represented class of farm size (38%) is the class 6-15 (Fig. 3), where are reared the 17% of the whole of the sows (Fig. 4). The farms of this class were the most investigated because of their importance for the area of study (small rural farms which are well established on the area). The 21% of the farms falls in the third class (16-50 sows) where are reared the 23% of the sows. The farms belonging to this class are the most interesting because they have the size enough to guarantee the self-sufficiency. At the same time this farms rear the animals with traditional techniques exploiting capitals and resources specific of the territory. The 51-100 and >100 classes are equally represented in the sample and have similar size in terms of number of sows (the smallest has 74 sows, the largest 104). The owners of these latter farms consider the livestock activity as an opportunity for capital investment and are able to exploit the favourable economic moment that has recently arisen in the rearing of Cinta Senese breed. In these farms the salami production is characterized by a high technologic level. In these two classes are reared almost the 50% of the sows (Fig. 4).

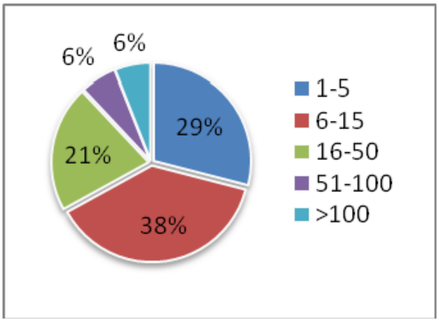

Fig. 3. Classes of farm size expressed as number of sows.

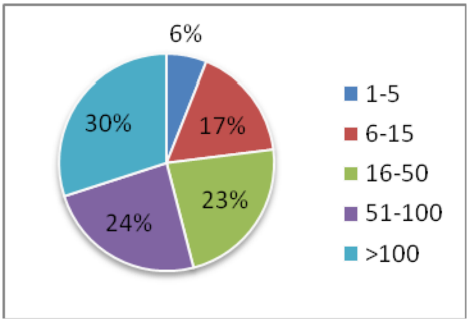

Fig. 4. Percentage of sows for each class of farm size.

The forest – when present – is used for grazing throughout the year by about 70% of farms (Fig. 5). The pasture – both natural and sown – respect to forest, is less exploited for breeding pigs, and approximately the 50% of farms never uses it for this purpose. Few, or never, farms use the stubble, or other categories such as olive groves, for grazing.

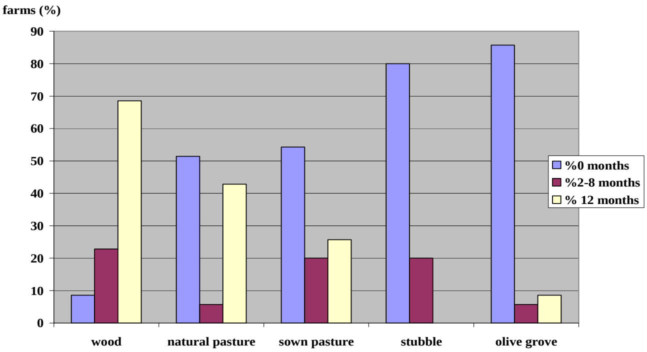

Fig. 5. Temporal exploitation of surface by farms (expressed as percentage of the total farms).

In Fig. 6 the 35 farms are listed according to the number of sows. The surface of the farms is not correlated with the number of animals raised, nor with the grazing area. There are farms with few animals and big land surface, as well as farms with many animals but with small land surface available for them, either for grazing or for crops dedicated to animal feeding. Farms with many animals, even when they have a large agricultural area available, use a very small area for the rearing of the pigs; this is because the Cinta Senese rearing has a very marginal role in this type of farms.

Figure 7 shows the degree of perception by the farmer of the environmental problems linked to the rearing of pigs, with particular reference to plant renewing and soil erosion. Over 50% of farmers do not find any kind of problem linked to outdoor pig farming, which is known to produce high impact. This result suggests that people minimizes the problem, this is for the lack of objective and encoded parameters for the measurement of damages.

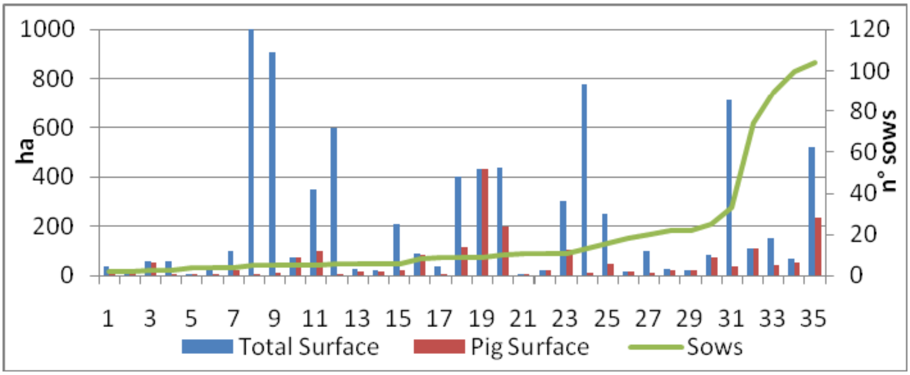

Fig. 6. Number of sows/ total farm surface and pastured farm surface by pigs.

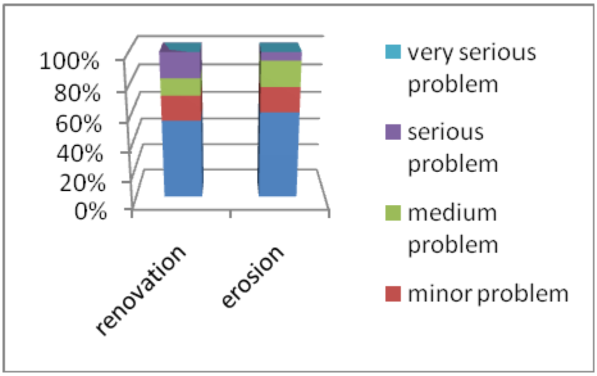

Fig. 7. Perception of environmental problems caused by the rearing of pigs.

The statistics of Fig. 8 have to be read considering that pigs are grazing on wood only in some periods of the year, particularly when the wood is able to provide its fruits for animal feeding. The 70% of farms grazed on wood the growing and fattening pigs for 12 months, while the 10% of them do not graze on wood these categories. The 55% of farms grazed the sows on wood for 12 months while the 30% of farms do not graze on wood these categories. This statistic raises serious doubts about the sustainability of natural resources because it seems that the forest is perceived as a "container" of pigs and not as a source of food supplements. It seems that only the 20% of the farms has a management according to with this principle.

Only the 20% of the breeders planes the reproductive activity and only in the 8% of the farms the sows have more than two births/year. In most cases (47%) the sows have two births/year, which is the more suited situation in the outdoor rearing system of Cinta Senese (Table 1). In many cases there are data that denote a very low reproductive activity; this is due, prevalently, to a bad management rather than to low fertility of animals. Indeed, taking into account the farming system adopted, the number of born and weaned piglets per litter is satisfactory; it is, on average, more than 7 and 6 piglets, respectively.

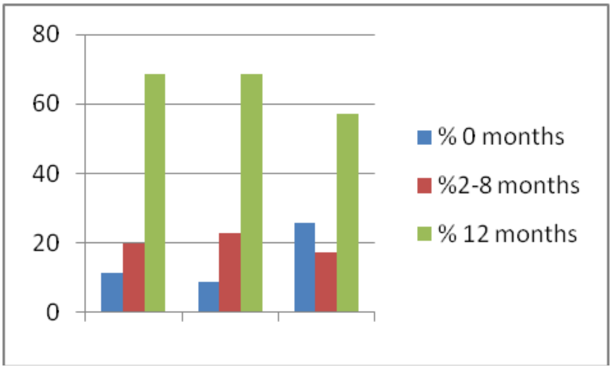

Fig. 8. Time of grazing on woods according to productive categories.

Table 1. Reproductive parameters

	Births/year/sow	Piglets/birth	Still piglets/birth	Weaned pigs/birth
Mean and s.d.	1.59 ± 0.66	7.31 ± 1.64	6.82 ± 1.48	6.16 ± 1.26
Median	2.00	7.00	6.75	6.00

All farms (except one) are equipped with structures where the sows deliver and suckle the litter (Fig. 9). Three farms are equipped with box for the birth similar to those used in intensive system. The 85% of farms use the consanguinity service provided by ANAS (Associazione Nazionale Allevatori Suini), the 83% of farms have no problems in giving maternity and paternity of the subjects.

The average age at which the piglets are weaned is 59.5 d (SD 24) and mode is 45 days (Fig. 10). All farms wean the pig above the minimum age scheduled for animal welfare (28 days); five farms wean within 36 days. There are many companies that wean the animals at age too high. The situation is detailed in the chart below.

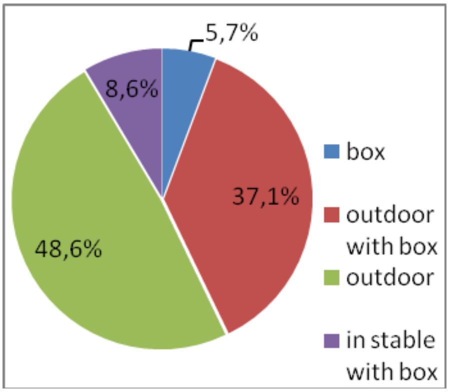

Fig. 9. Type of births adopted by farms.

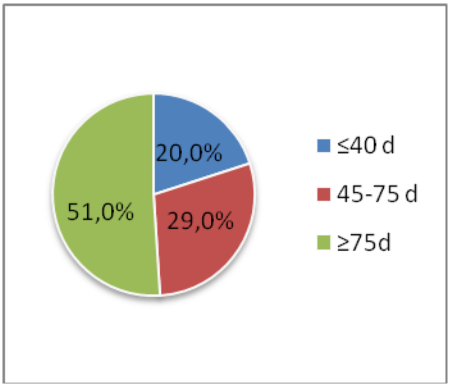

Fig. 10. Weaning age (days).

Figure 11 describes, for each farm, the relationship between weight and age of slaughter. It show that the final product is characterized by great heterogeneity which has a negative impact on the quality of the final products. The slaughter weight, which should be around 150 kg (130 to 170), can be reached at 12-15 months with diets characterized by medium-high energy level. This weight can be achieved in 18-21 months with diets characterized by medium-low energy level, mainly on pasture (in figure 11 these two situations are represented by the blue and red ovals respectively).

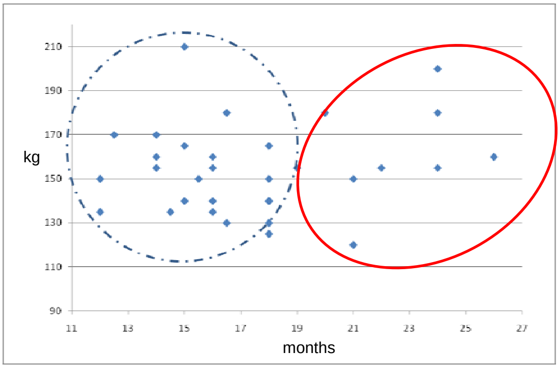

Fig. 11. Slaughter age/live weight for each farm.

The pigs, both sold live and after slaughter, are mostly used for salami production. However, 27 % of farms sales some animals at the age of weaning; these pigs will be fattened by others. The 8% of farms sales light animals to make whole roasted pig.

Regarding to products market, Fig. 12 reports the final destination of carcasses; it's interesting to report that over 40% of farms use a single sale channel, two sale channels for 38% of farms, 22% of farms distributes its production in 3 or more sectors.

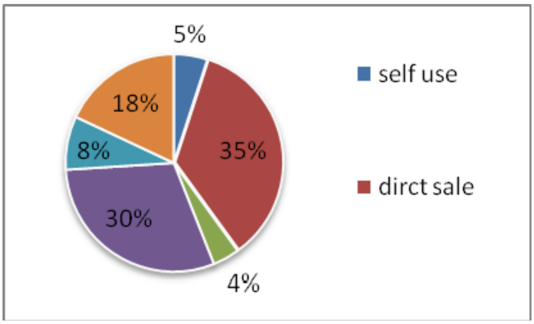

Fig. 12. Products market.

Large part of the farmers interviewed (75%) stated to delegate to third parties the processing of their products because they do not have the facilities for carcass dissection and seasoning of meat. Two out of 8 processing companies analyzed belong to farmers as a terminal structure of the production cycle and they are used almost exclusively for personal use or otherwise for the production of their products. The other 6 are professional processing companies. Among these one is an industrial-type and five are of medium size. The products obtained from Cinta Senese pig are various: cured ham, salami, bacon, *coppa*, *soppressata*, *finocchiona*, shoulder and belly. Only 7 companies produce Salami and *coppa* whereas lard is produced by 5 companies and the ham is processed by all the companies. Two processing plants prefer the winter production while the other processing plants are active all along the year. One of them uses traditional drying chamber while all the others have been equipped with cold air cooling system. Depending on the product, the surveyed companies employ varying amounts of additives. The additives are used in sausage and in the *coppa* from 44% of the companies, in lard from a single company and ham from 2 companies. Twenty-five percent of the processing plants sell the products abroad as well as in the national and local markets, such percentage rises to 50% if one considers only the ham. The production is almost exclusively destined for sales with the exception of the smallest and traditional company which also produces for private use. All the companies selling products as a whole or as pieces and only the biggest company also produces sliced products commercialized in vacuum package.

Salami: For the production of salami, two companies prefer to add pork from improved breeds to the mixture in order to reduce problems during maturation, which may occur due to excessive amounts of fat and to its poor quality. The processing technique is: the lean raw material comes from the shoulder whereas the fat comes from the belly and the lard. One company also uses the loin and the ham. Five companies add a small dose of garlic and one of them also wine and hot pepper.

***Coppa* or *Capocollo*:** It is a product that requires a smaller work than the salami. Besides the salt is added a small amount of garlic (premixed with the salt itself) while two companies also add the fennel. Three companies covered all the product with salt. All companies washed the product after the period of curing whereas 60% of them use a wrapping package. Five out of 7 companies producing *coppa* carry out the drying step while the other two move the product directly in the cells of aging.

Lard: The lard is the less represented product in the surveyed companies (only 5), perhaps because in many cases it is used for the production of sausages. Almost all the companies cover the cut with salt, 4 companies add also pepper and garlic while only 3 companies add other spices (rosemary, bay leaves, juniper, nutmeg). Two companies made also the intermediate stages of washing and drying after the salting period whereas two other companies put the product directly in the maturation room after salting; one company sell the product immediately after the salting period.

Ham: As regard the ham, the fresh leg undergoes to an average trimming in 5 plants, to reduced trimming in 2 and to high trimming in one plant. The ham is with bone in all the plants. The thighs are fully covered with salt in 4 plants while in other 3 plants only a partial coverage of salt is provided. All the plants add pepper and 6 out of 7 include garlic. Five plants perform manual washing and 4 after the wash rest the thighs for a while. The drying step is carried out by 5 plants while the other two go directly to the curing phase. The end of the curing time is time-depending and only 3 plants control sometimes the weight of the final product (Table 2).

Table 2. Parameters of ham making

	Weight kg	Days of salting	Resting period	Drying			Curing		
				Days	T °C	Humidity %	Months	T °C	Humidity %
Mean	14.5	25.5	70	42.33	14.6	72	10.5	15.8	77.5
Median	14	21	60	60	14	75	11	14	77.5
Mode	13	21	60	60	14	75	-	14	80
s.d.	2.06	17.16	17.32	30.60	5.55	7.58	3.42	3.49	2.89

IV – Conclusions

The results of this survey can evidence some strengths and weaknesses points of this breed:

Strengths. The main strength is the territorial identity of Cinta Senese rearing and product, which is particularly excited by the name and label of Tuscany. This advantage is palpable and well recognized by the farmers who rely on this link for the marketing of products. The first farmers / entrepreneurs followed this line of propaganda to present the product of Cinta Senese on important international niche markets, thus starting the promotion of the breed, which still ensures the excellence of the product and its brand recognition. The support of public organisms, the interest of scientific research and the activities of the newborn Consortium for the PDO have helped the breed to earn its commercial reputation, thus eliminating completely the risk of extinction. The main strength of the production of salami of Cinta Senese is the good image acquired by the breed and it has an impact in another positive parameter: the selling price. Further strength is definitely the raw material from which it originates. The quality of meat and especially of the fat component, is influenced by the acidic composition. The acidic composition and the quality of fat are linked to the genetic of the breed but they could be modulated with specific plans of nutrition also influenced by the rearing systems adopted. In fact it is well known that the extensive farming, if correctly exploits, plant resources and especially the berries, may lead to products with peculiar organoleptic characteristics coming from a different acidic composition in turns due to specific fatty acids of the diet. From the perspective of technological transformation, they don't appear particular strengths, or at least not in all the surveyed plants; the only peculiarity is the use of garlic for all the products and, in one plant, the use of traditional not air-conditioned rooms.

Weaknesses. The strong interaction rearing-environment does not always protect the latter. The wood in particular seems the most suffering component and environmental damage caused by the indiscriminate rearing in some areas could also have some serious long-period consequences. Breeders of Cinta Senese have very different extraction often displaying a careless approach, especially in the early stages. This has led to a strong variability of behaviour that influenced the characteristics of products, with the risk that those of low quality can compromise the reputation of the entire system. The strong individualism of Tuscan breeders that can be mitigated only by strengthening role and actions of the Consortium for the PDO protection. Regarding the technical and marketing strategies, the weaknesses are: the lack of common facilities; the distance, for some farms, from slaughterhouses and processors, which leads to increased production costs; the great variability in the quality of the raw materials due to different livestock management, especially in terms of feeding and age and slaughter weight of the animals. Such variability is not declared in the final products, so the consumer finds under the same typology of products different quality with prices not always proportional. Even the curing process is not standardized, and for some aspects is "top secret" and this also affects the typology of the final products. Another gap, not yet overwhelmed, is the production and marketing typologies that are not yet standardized; the demand/supply ratio is often unstable since the chain is not well structured.

Acknowledgements

The authors wish to acknowledge the financial support received from ARSIA and Ente Cassa di Risparmio di Firenze for its support in purchasing SAS licence.

Evaluation of forest damage derived from the rearing of Apulo-Calabrese pig

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Abstract. The Apulo-Calabrese is one of the five Italian native pig breeds, officially recognized. At the moment it is reared in 35 farms, 29 of them are localised in the Calabria region. The aim of this work was to assess animal management with particular attention to the forest grazing. The survey involved 20 farms and in 4 of them the forest damages in two seasons (summer and autumn) were evaluated. 80% of farms pastured forest all the year, 75% supplied concentrate to the animals irrespectively of pasture. 67% of farms pastured all the productive categories. The damage to trees from barking and root uncovering was higher in summer and decreased with the increased distance from the shelters. The slope of the soil increased the damage by uncovering (0.2% per degree). The most uncovered tree species were chestnut and phillyrea, while these most barked were holm, oak and phillyrea. Soil and litter were heavily damaged near the shelters (30.29% and 52.47%, respectively). Soil trampling and excavation damage were found in 31% and 36.5% of the experimental areas, respectively. Finally, the greatest damage was observed in the farms with the highest animal loading.

Keywords. Local pig – Apulo-Calabrese breed – Pasture in forest – Forest damage.

Evaluation des dégâts forestiers causés par les troupeaux de porcs de race Apulo-Calabrese

Résumé. *La race porcine Apulo-Calabrese est une des cinq races porcines indigènes italiennes officiellement reconnues, et actuellement élevée dans 35 exploitations, dont 29 dans la région de Calabre. Le présent document vise à évaluer la gestion des animaux en mettant l'accent sur le pâturage dans la forêt. L'enquête a porté sur 20 exploitations et 4 d'entre elles ont évalué les dégâts causés au bois à deux saisons (été et automne). 80% des exploitations font brouter la forêt tout au long de l'année; de toute façon 75% distribuent du concentré. 67% des exploitations agricoles font pâturer toutes les catégories d'animaux. Les dégâts causés par l'écorçage et par le creusement autour des racines des plantes ligneuses sont plus élevés en été et diminuent avec la distance de l'abri. La pente du sol augmente les dégâts du creusement (0,2% par degré). Les arbres les plus écorcés sont: châtaignier et phillyrea ; les plus creusés: chêne vert et phillyrea. Les sols et la litière sont fortement endommagés près des abris (30,29 et 52,47%). Le piétinage du sol a été observé dans 31% des cas et le creusement du sol dans 36,5% des cas. Les dégâts les plus importants ont été observés avec le chargement animal plus élevé.*

Mots-clés. *Porc rustique – Race Apulo-Calabrese – Pâturage dans la forêt – Dégâts aux forêts.*

I – Introduction

In recent decades, the scientific community and institutions have recognized the need to preserve animal germoplasm to limit the decline in genetic variability caused by the selection and the use of few "improved" breeds. With this perspective it could be inserted the recovery and preservation, carried out in Calabria, of the Apulo-Calabrese breed. In farms that raise this breed, grazing in the forest is often implemented. This practice should be done with extreme caution because the pigs, given their ethology, can alter the soil as well as damage the plant component and affect the hydro geological function (Grifoni and Gonnelli, 2009). The purpose of this study was to describe farms that rear Apulo-Calabrese pigs in province of Cosenza (Calabria), and to estimate the impact of grazing on the forest.

II – Materials and methods

Description of herds: The survey involved 20 farms. The information, collected on special sheets, were: farm size, land use, herd consistency and feeding management. Averages, ds and frequency of classes were calculated.

Impact on forest: The measurements to assess the entity of the impact on the forest were conducted in four herds into two periods of the year, summer and autumn. The method used for the assessment of damages was essentially that proposed by Grifoni *et al*. (2007). For all parcels, slope, distance from the shelter and loading (number of animals / ha) were also detected. In this study only some of the parameters of damage are analyzed: uncovering of the roots and barking of woody plants, litter movement, alteration in the humus layer and soil trampling and excavation. Data on damages were submitted to analysis of covariance (SAS, 2003),using, as discrete factors, season (summer, autumn), distance from shelter (four classes: in shelter, near, intermediate, distant), tree species (*Quercus ilex, Quercus cerris, Quercus pubescens, Castanea sativa, Phillyrea latifolia*) and, as continuous factors, animal loading and ground slope.

III – Results

Description of herds: The farms are ranked by total surface size and Table 1 shows the descriptive statistics.

Table 1. Means of farm features according to dimensional classes

	Farm surface		
	< 20 ha	**21- 100 ha**	**> 100 ha**
Number of farms	13	6	1
Total farm surface (ha)	6.9	35.2	420
Farm surface for pig (%)	62	34	5
Number of pigs	17.2	60.5	42
Pigs/wood surface (n/ha)	10.9	11.4	2.1

Most farms (65%) are small (<20 ha), with lower extension than those who raise other rustic Italian breeds, such as Cinta Senese (Bonanzinga and Nardi, 2007) and Nero Siciliano (Costanza *et al.*, 1990), and this fact is confirmed by a recent survey by Nicolosi *et al*. (2009). Only one farm has surface larger than 100 ha. The stratification by size shows that midsize farms have, on average, the highest number of animals.

The number of animals reared per farm (Table 2) shows an average of 31.45, with wide variability (± 45.39). Only one herd has no breeding animals and it raises exclusively for fattening; other herds raise 14 sows and two boars, on average. The values are higher than those observed by D'Ancona (2001) for Nero Siciliano (8.1 sows and 1.5 boars) and Bonanzinga and Nardi (2007) for Cinta Senese (9.5 sows and 1.6 boars).

Table 2. Average consistency of farms according the animal category

	Sows	Boars	Gilts	Small boars	Total
Average	14.35	2.3	8.35	5.95	31.45
S.d.	27.43	3.26	11.81	9.62	45.39

As feeding management is concerned, the 80% of farms holds wood areas where grazing takes place throughout the year while the 20% suspends the pasture during the summer (June-September). No farm has meadows and pastures, unlike the situation of breeding of Cinta Senese in Tuscany (Giuliotti and Ferrini, 2004).

In 67% of farms all categories of pigs are sent to pasture while, in the other farms, the pasture is dedicated exclusively to the breeding animals. Regarding the animal loading, defined as the number of animals per hectare of grazed area, it is resulted an average of 7.7 heads with a large variability (SD ± 5.9). However, in every herd feeding supplement is adopted using either commercial mixtures, or concentrates of farm origin, regardless of grazing; in fact the feed administration occurs throughout all the year in 75% of cases (daily dose of 2.5% of PV), while the remaining 25% of farmers suspends the feeding in autumn, to coincide with the fall of chestnuts and acorns.

Impact on forest: Table 3 shows that in summer the percentage of barked plants was much greater than in the autumn and this may be attributable to the presence, in the autumn of acorn. For uncovering of roots differences do not reach statistical significance.

Table 3. Effect of season and distance from shelter on trees damage (estimated to average loading of 2.92 pig/ha and ground slope of 28.84%)

	Season		Distance			
	E	**A**	**On shelter**	**Near**	**Intermediate**	**Distant**
% Uncovered roots	25.95	18.27	63.22 a	17.30 b	5.80 c	2.13 c
% Barked plants	26.10 a	9.14 b	49.93 a	15.10 b	3.60 c	1.85 c

Different letters for P<0.05.

The two types of damage decreased significantly with increasing of distance from the point of shelter. Finally, the percentage of uncovered roots was found to be influenced by the slope of the soil (+ 0.20 percentage points for each degree of slope, data not tabulated).

The two types of damage were analyzed also for the effect of the different tree species (Table 4). Table shows that for uncovering there were not significant differences, although *Phillyrea latifolia* and *Castanea sativa* seemed the most damaged. For barking, on the contrary, *Quercus ilex* and *Phillyrea latifolia* were more damaged while *Quercus pubescens* (P<0.05) was the species that suffered less damage.

Alteration of litter and humus (Table 5) was very influenced by the distance from the animal shelter, because the damage decreased progressively with the distance. The humus was also damaged by the increase of loading (P<0.05) but not by the ground slope (data not tabulated). The soil trampling (data not tabulated) were detected in 31% of cases and were evaluated for type light, medium and high in 48, 9 and 43% of cases, respectively. The damage from excavation were observed in 36.5% of cases, with low entity in 18.5%, medium in 48% and strong in 33.5% of cases.

Table 4. Effect of tree species on trees damage (estimated to average loading of 2.92 pig/ha and ground slope of 28.84%)

	Castanea sativa	***Quercus cerris***	***Quercus pubescens***	***Quercus ilex***	***Phillyrea latifolia***	**DSR**
%Uncovered roots	25.38	18.44	20.65	19.32	24.60	12.88
% Barked plants	15.90	19.20	13.98 a	27.83 b	26.92 b	13.25

Different letters for P<0.05

Table 5. Effect of season and distance from shelter on the damage on litter and humus

Category	Season		Distance from shelter				
	E	A	On shelter	Near	Intermediate	Distant	DSR
% Altered litter	61.20	54.86	96.84 a	52.47 b	48.10 cb	34.73 c	26.56
% Altered humus	38.10	34.11	84.00 a	30.29 b	15.06 b	15.10 b	26.80

Different letters for P<0.05.

IV – Conclusions

The survey shows that the breeding of Apulo-Calabrese pig is implemented mainly in small farm and rarely ignores the presence of the forest, according to the vocation of the breed to be reared outdoors. The survey shows, however, that the grazing is little managed and pigs remain in woods throughout the year, in almost all cases. The loadings are high and food requirements of animals are supported by high dose of concentrates, irrespective from the pasture. The relation between loading and damages must be explored. Looking at the damage that Apulo-Calabrese pig determined both on the forest plants and on the soil, it appears that the impact was substantial, especially in summer and in the proximity of shelters and it became very serious within the shelter. All tree species suffered barking and root uncovering, the latter increasing with ground slope. This suggests the need to organize the shelters outside the forest. In conclusion, in our point of view, to avoid waste, reduce environmental impact and ensure animal welfare and product quality it must be implemented a system that: (i) turns grazing surface; (ii) limits grazing in the forest to the period in which it provides food (acorn and chestnut); (iii) provides other types of pasture, as meadows; (iv) administrates diet according to the needs of animals and the availability of pasture resources.

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Effectiveness of immunocastration in adult boars

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Abstract. Immunocastration (IC) of pigs during their finishing stage avoids the traumatic castration surgery (Q) still practiced in many countries to prevent boar taint in pork. We have not found reports on IC of culled adult boars. In this study we evaluated boar taint, spermatogenesis, and weight loss of adult boars after castration by IC versus Q. A total of 21 boars were used (age: 29 months). The IC boars (n: 12) were injected with Innosure® (Pfizer Inc), and repeated four weeks later. The Q pigs (n: 9) were castrated when the IC were first injected. Both groups were monthly weighted and slaughtered five weeks after the second injection. Testes of IC slaughtered boars were sampled and compared with the Q group. Odor panels were conducted to test all carcasses and pork. The Q boars lost 0.172 kg BW/animal/day, while IC boars did not lose weight (P=0.016). None of the boars (IC or Q) resulted in tainted pork. The IC testes had lower spermatogenesis compared to Q. It is concluded that IC effectively prevented boar taint through testicle atrophy, and it also resulted in no weight loss after castration.

Keywords. Immunocastration – Immunological castration – Immunocontraception – Boar taint – Sex odour – Mature boar.

Efficacité de l'immunocastration des verrats adultes

Résumé. *L'immunocastration (IC) des porcs pendant leur phase de finition permet d'éviter la castration traumatique (Q) encore pratiquée dans de nombreux pays pour éviter l'odeur de verrat dans la viande du porc. Nous n'avons pas trouvé de rapports sur l'IC de verrats de réforme. Dans cette étude nous avons évalué la spermatogenèse, l'odeur de verrat, et la perte de poids des mâles adultes après la castration par IC versus Q. Un total de 21 verrats ont été utilisés (âge: 29 mois). Les verrats IC (n: 12) ont été injectés avec Innosure ® (Pfizer Inc), et le traitement a été répété quatre semaines plus tard. Les verrats Q (n: 9) ont été castrés au même moment où les IC ont été injectés. Les deux groupes ont été pesés deux fois et abattus cinq semaines après la deuxième injection du groupe IC. Les testicules des verrats IC abattus ont été échantillonnés et comparés à ceux du groupe Q. Des panels relatifs aux odeurs ont été réalisés afin de tester toutes les carcasses et les échantillons de viande. Les verrats Q ont perdu 0,172 kg de poids corporel/animal/jour, tandis que les verrats IC n'ont pas perdu de poids (P = 0,016). Dans aucun des groupes on n'a trouvé (IC ou Q) de contamination d'odeur. Les testicules IC ont présenté une spermatogenèse diminuée comparés aux Q. Nous en concluons que l'immunocastration est capable de diminuer efficacement l'odeur de verrat par une atrophie des testicules; de plus, aucune perte de poids après la castration du groupe IC n'a été détectée.*

Mots-clés. *Immunocastration – Castration immunologique – Immunocontraception – Odeur de verrat – Odeur sexuelle – Verrat.*

I – Introduction

It is well known that non-castrated fattening pigs perform better than barrows (Harding, 1993). Nevertheless, pork from non-castrated boars may present an unpleasant odor and taste to the consumer. This is due to the combined effect of androstenone derivatives and skatole deposited in the meat (Bonneau, 1982; Brooks and Pearson, 1986; Xue and Dial, 1997). Immunocastration (IC) is an immunological castration method which is currently used worldwide to castrate pigs at the end of their fattening stage. The IC method works as a vaccine, stimulating the immune system to produce antibodies against the gonadotropin-releasing hormone (GnRF), ultimately inhibiting the generation of androstenone (Ferro, 2002). Immunocastration is an alternative to the usually traumatic surgical castration still in use in many countries (Prunier *et al*., 2006). Besides being regarded as animal-friendly, IC is also beneficial to the producer of grow-fattening pigs, considering that IC pigs grow leaner, have higher weight gain and better feed conversion ratio compared to pigs surgically castrated early in life (Dunshea *et al*., 2001; Schmoll *et al*., 2009). The performance advantages of a late castration are related to a longer exposition to androgens (Xue *et al*., 1995). We are not aware of studies that have evaluated the effectiveness of IC in adult boars once they have completed their productive life in the breeding farm and should be castrated before slaughtering. The main objective of this study was to evaluate the effect of IC on boar taint of adult boars.

II – Materials and methods

The study was conducted in three commercial pig farms located in Antioquia, Colombia. A total of 21 boars were used. The average age of the boars was 29 months (range: 26 to 36 months). To start the trial, twelve boars received a 2 ml subcutaneous injection of Innosure® (Pfizer Animal Health, Parkville, Australia) in the neck, close to the base of the ear. This group (IC) received a second injection four weeks later. The day of the first injection, another group of boars was surgically castrated. Castration of this second group (Q) was conducted with the same technique in the three farms. From trial start until pig slaughtering the boars were fed the same feed as before (2 kg/boar/day). Boars were weighed on three occasions: at the starting of the trial (defined by the first vaccination of IC or the surgical castration of Q), at the second vaccination (four weeks later), and at the end of the trial (five weeks later). Testes of IC slaughtered boars were sampled and spermatogenesis was compared with the testes taken from the Q group. Fifty observations (seminiferous tubules) per testicle were conducted to assess spermatogenesis scores, using the procedure reported by Johnsen (1970) and modified by Peters *et al*. (2000). Incidence of objectionable odor was assessed by odor panels conducted to test all boars. This was done by sniffing the hot carcasses right after slaughter, and then by sniffing a sample of pork previously warmed in a water bath, according to the method described by Judge *et al*. (1990).

III – Results and conclusions

The vaccine injection was well tolerated by the boars. No observable site reactions were detected at the time of slaughter. In general, testes size was notoriously reduced in the IC boars. Although the size variation was not measured, a picture is presented to give an idea of the difference in testis size at the end of the nine weeks of the trial (Fig. 1). Figure 2 reflects spermatogenesis scores. Scores from one to seven indicate lower production of sperm cells. Larger scores (e.g. greater than seven) imply normal spermatogenesis. The IC testes had lower scores compared to Q (seven versus 10, respectively).

Table 1 shows the body weight changes observed between treatments for the three weighing intervals (first to second, second to third, and first to third weighing). For the third weighing interval, which determines the weigh change for the overall trial, the Q boars had lost 0.172 kg BW/animal/day, while IC boars did not lose weight (P=0.016).

According to the odor panel results, none of the boars (IC or Q) resulted in tainted pork. It is concluded that IC effectively prevented boar taint through testicle atrophy, and it also resulted in no weight loss after castration.

Acknowledgements

The authors want to thank the Colombian Instituto Nacional de Vigilancia de Medicamentos y Alimentos (INVIMA) for the supervision of the odour panels. This research was supported by the University of Antioquia and Alimentos Cárnicos SSA.

Fig. 1. **Two testes longitudinally cut from a surgically castrated (Q) boar (left and center), and a testis from an IC boar. Note the size difference.**

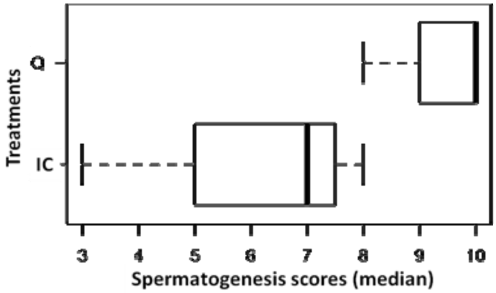

Fig. 2. **Median spermatogenesis level for immunocastrated (IC) versus surgically castrated (Q) adult culled boars.**

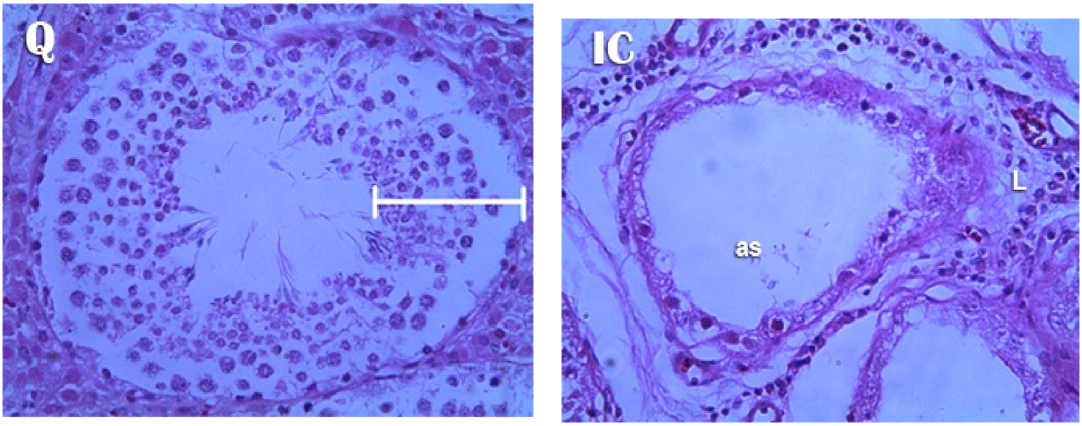

Fig. 3. **Comparative sections through the testis of a surgically castrated (Q) and an immunocastrated (IC) boar following H.E. staining (400x magnification). Note the presence of all sperm stages in the Q testis, compared with azoospermia (as), depleted, and degenerated Leydig cells (L) in the IC testis. The magnification is the same for both treatments.**

Table 1. Body weight changes (kg/day) between weightings for immunocastrated (IC) versus surgically castrated (Q) adult culled boars

Treatment	Period of time between weightings	Ave. body weight change (k/d)*	T test**		
			t- value	DF	P-value
IC	1st to 2nd	0.079 ± 0.06	a 1.410	19	0.1748
IC	2nd to 3rd	-0.026 ± 0.05	a -0.482	19	0.6354
IC	1st to 3rd	0.053 ± 0.06	a 0.940	19	0.3590
Q	1st to 2nd	-0.279 ± 0.07	a -4.29	19	0.0004
Q	2nd to 3rd	0.107 ± 0.06	b 1.69	19	0.1084
Q	1st to 3rd	-172 ± 0.07	a -2.65	19	0.0160

*Average plus or minus the standard error. **T-test corresponding to each line. DF: Degrees of freedom.

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Proposal of sustainable management of pig farming on pastures in Tuscany

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Abstract. It is quite recent in Tuscany the rediscovery of rearing on pastures of rustic pig breeds. The utilization of the pastures is often implemented without a specific skill of the farmer and this often leads to feeding errors exceeding the carrying capacity and wasting the feed; this situation has negative impact on the ecosystem, the animal welfare, the quality of products and on the farm management. This paper is aimed to propose a practical scheme of Cinta Senese rearing on pastures focused on the use of wood as a factor characterizing the quality of production. The example (module to be replicated in case of larger farms) is a farm, with 9 sows producing progeny two times per year. Feeding plans, reported as graphs, provide forage chains suitable for the rearing area of Cinta Senese as well as the amount of feed (kg) to be used daily per animal during the entire growth-fattening phase. Length of rearing on pastures and feeding plan are function of season of birth, growth intensity and expected availability of pasture that will affect the final product quality.

Keywords. Cinta Senese breed – Rearing on pastures – Reproductive timetable – Feeding cycle.

Proposition de gestion durable de l'élevage porcin sur pâturages en Toscane

Résumé. En Toscane l'élevage en plein air des porcs des races rustiques a récemment été redécouvert. L'utilisation des ressources pastorales est souvent faite sans que l'éleveur soit un expert ce qui conduit souvent à des erreurs de rationnement avec surcharge ou déchets; l'impact négatif retombe sur l'écosystème, le bien-être animal, la qualité des produits et la gestion économique des entreprises. Cet article propose un diagramme d'application du pâturage pour l'élevage de la Cinta Senese, où l'accent est mis sur l'utilisation du bois comme élément caractérisant la qualité de la production. L'exemple (le module doit être répliqué dans le cas d'une grande entreprise) est un type d'exploitation agricole, avec neuf truies qui donnent deux portées par an. Les plans d'alimentation offerts, sous forme de graphiques, montrent les chaînes d'alimentation qui peuvent être proposées pour l'élevage de la Cinta Senese et montrent la quantité de nourriture à utiliser quotidiennement par tête, tout au long de la phase de croissance et d'engraissement. La durée de l'élevage et du plan alimentaire dépendent de la saison de naissance, de l'intensité de la croissance et de la disponibilité des pâturages, qui aura une incidence sur la qualité du produit final.

Mots-clés. Cinta Senese – Élevage extensif – Calendrier reproductif – Chaîne d'alimentation.

I – Introduction

The extensive production systems, in which pigs graze outdoors for much of their life, have an ancient tradition in many European countries. From the middle of the last century, economic and social reasons required to increase the efficiency of production and to contain production costs. Consequently systems of indoor intensive farming were established. The recent resurgence of outdoor farming was determined by a combination of factors: (i) the low value of the land in some marginalized areas; (ii) the increased cost of structures, management and equipment; (iii) the implementation of strict regulations about storage and distribution of manure; (iv) the breeder needs to answer the demand for "genuine" products by consumers; and (v) the awareness of the need for farming systems best suited to animal welfare and eco-friendly.

The outdoor farming with the use of grazing in the woods most of the year, however, can produce serious risk of degradation of this fragile ecosystem with a negative impact on the environment and on the animals (Franci *et al*. 2004; Acciaioli *et al*. 2010) The aim of this paper is to propose a breeding scheme, calibrated on Cinta Senese breed, that through precise management of reproductive animals, allowing a diverse use of available resources, and allow to enhance the production while safeguarding the environment.

II – Materials and methods

The work consists in the formulation of a management scheme, settled to the actual situation of Cinta Senese breed in Tuscany (Bonanzinga *et al*., 2007). The example is calibrated on the average number of animals found for Cinta Senese farms equal to 9 sows, divided into 3 groups (A, B and C) which foreseen scalar calving (every two months) with 2 calving per year (1 and 2) for each sow (Table 1).

Table 1. Calving timetable

	Jan	Feb	Mar	Apr	May	Jun	Jul	Aug	Sep	Oct	Nov	Dec
I° calving		A 1		B 1		C 1						
II° calving								A 2		B 2		C 2

Considering that: (i) it is not economically viable to prolong the breeding of animals over 18-19 months; (ii) the final product quality is essentially due to the feed received during the finishing period; (iii) the acorn and the pasture in wood are the elements that characterize this production; and (iv) the seasonality of forest resource does not allow to use it for the animals born during all periods of the year; it is proposed a scheme (Table 2) that allows to use the acorn in the finishing period by the largest number of pigs.

Table 2. Timetable of pasture in wood (only animals in the finishing phase, the groups are the maternal ones)

	Jan	Feb	Mar	Apr	May	Jun	Jul	Aug	Sep	Oct	Nov	Dec
I° calving mod. A		**A 1** **12 m**										
I° calving mod. A				**B 1** **12 m**								
I° calving mod. B									**B 1** **17 m**			
I° calving mod. B										**C 1** **16 m**		
II° calving mod. C										**A 2** **14 m**		
II° calving mod. C										**B 2** **12 m**		
II° calving mod. C												**C 2** **12 m**

Finishing on wood | *Finishing not on wood*

To achieve the use of the scheme a more or less rapid average daily gain has to be encouraged according to time of birth in order to reach at least 110 kg at the start of grazing in wood for all the animals.

Feeding of the animals follow different patterns, which have been identified and summarized in 3 models:

- Model A, on animals that do not use the wood: the growth will preferably be rapid because extending beyond 15 months at this stage would not have reasons neither economic nor for the quality of products;

- Model B, on animals that will undergo to slow growth waiting for the production of acorns in the fall in which they will have an age of about 16-17 months;

- Model C, relative to animals subjected to rapid growth so they will reach a reasonable weight before the introduction on wood pasture, despite their young age (12-14 months).

III – Results

The analysis of Table 2 shows that:

- Individuals born in February (group A1), do not use the wood for the fattening phase and they could be slaughtered from 15 months of age (Model A);

- Animals born in April (group B1) can be finished without wood (model A) or be slaughtered after grazing on wood (model B) extending the period of growth;

- All the animals born in June (Group C1) may be subjected to fattening in wood and they will be slaughtered at about 19 months of age (Model B);

- All the animals born in the second calving (A2, B2, C2) may be finished on wood and they will be slaughtered at an age between 15 and 17 months. (Model C).

The proposed scheme, which expects to produce some batches of animals without grazing in wood has some positive aspects including the fact that it has released by seasonality at least for a part of the production and it can also increase the farm production containing the load on the forest. In this case the feeding has to respond to qualitative criteria such as to obtain such a product that can be distinguished from products of intensive farming. This solution is particularly desirable in the years when acorn production is poor.

Figures 1, 2 and 3 reported examples of food plans, linked to the "models" proposed, and they show the amount of food used daily by each animal for the entire growing-fattening phase, expressed in kg of food such as (fresh weight). The use of wood pasture and grazing on grass and stubble is foreseen. The integration with concentrates is aimed at balancing the rations. Rations take into account the growth rate of the animals and the food present in the different periods of the year.

The variability of pasture resources in different years, does not allow the preparation of rigid plans and it is even harder to determine in advance the load; the use of all farm resources has to be done by changing from time to time the load and the rotation of the plots on basis of their productivity, and this will be the major objective for sustainable management, not forgetting to use the most valuable resources (acorn and chestnut) for finishing by transferring this value in the processed products in order to valorize them (Pugliese *et al*. 2007).

Figure 4 shows the food consumption for each of the proposed models, expressed in dry matter, for the entire cycle of rearing; it is evident that model C allows the use of more food coming from wood.

Table 3 finally reports the daily intakes and the total conversion indexes of the growth and fattening phases (from 20 to 140 kg), based on the proposed models and as a comparison it was included a column with data for fattening with only concentrates always reared outdoors with slow growth rate (370 g/d) (AA.VV., 2004). The conversion index of dry matter (kg food consumed/kg of daily gain obtained) is better when the farming systems are short due to the decrease of the overall cost for the maintenance of the animals.

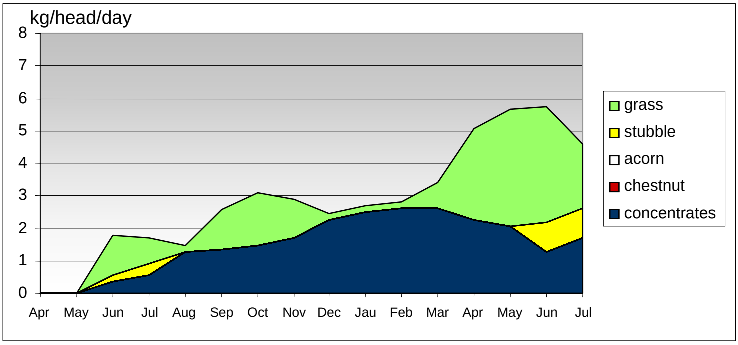

Fig. 1. Feed intake (kg of fresh weight) MODEL A: pig born in spring and slaughtered at 15 months of age, with normal growth and without finishing in the wood.

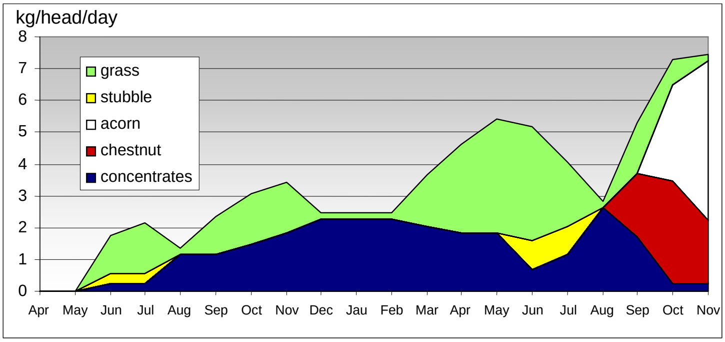

Fig. 2. Feed intake (kg of fresh weight). MODEL B: pig born in the spring slaughtered at 18 months of age, with moderate growth and finishing on pasture in wood.

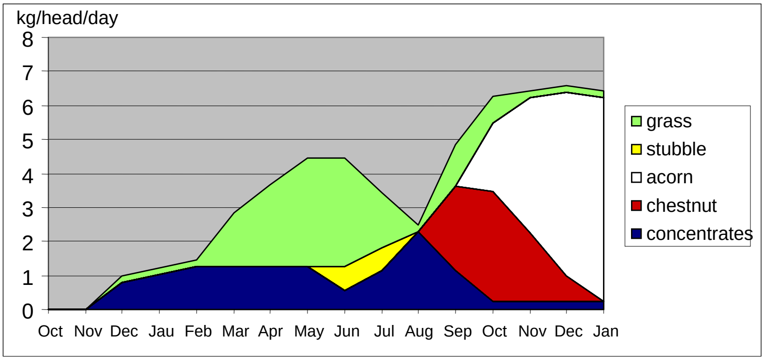

Fig. 3. Feed intake (kg of fresh weight) MODEL C: pig born in autumn and slaughtered at 15 months of age, with normal growth and finishing with grazing in wood.

IV – Conclusions

The knowledge and considerations reported here show that, under wild and semi-wild breeding it is difficult to both formulate plans and organize complex suitable feeding plans. In fact food availability will also change significantly over the year and affect the timing and the pattern of body growth of animals. The Italian territorial reality, but also the breeding area of Cinta Senese is diverse and therefore we can only propose some solutions (Models) that surely can not reflect all the situations. For proper farm management it s also required to implement a consistent control of animals to confirm the correctness of the feeding plan adopted (body condition, health and behavior), and also the environment to ensure the balance of the grazing system.

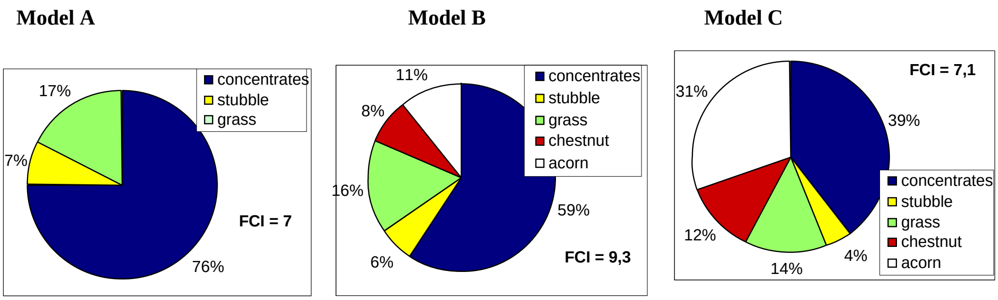

Fig. 4. Percentage of dry matter ingested in the 3 different systems of feeding offered during the whole growing-fattening period.

Table 3. Total consumption per animal to ensure an increase of 120 kg (20 to 140 kg) for each feeding plan considered

	Model A	Model B	Model C	Only concentrates
Feedstuff (kg t.q.)	784	753	474	922
Cereals on stubbles (kg t.q.)	76	72	41	-
Grass (kg t.q.)	600	726	522	-
Chestnut (kg t.q.)	-	217	210	-
Acorn (kg t.q.)	-	228	516	-
Total intake of DM (kg)	903	1108	925	811
Days	420	540	420	420
Conversion index of DM (FCI)	7	9,3	7,1	6,7 *

*with average daily gain of 370 g.

Acknowledgements

The authors wish to acknowledge the financial support received from ARSIA.

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The Nebrodi Black Pig: description of the regional situation with particular attention to the environment and the techniques and types of farming

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Abstract. The Black Pig, bred in the Nebrodi, represents a familiar cultural heritage passed from previous generations. Although the breeding of modern breeds represents significant economic benefits, it is thanks to interventions of Department for Agriculture and Forestry of Sicily Region that it was possible to avoid the loss of the genetic heritage and the extinction. The peculiar sensory characteristics of the meat and specifically of the fat, combined with the typical of seasoning, make that the products are particularly appreciated by consumers. The objective of this search, that is part of a wider context, is to estimate the spread and the numerical abundance of the agricultural companies, in the productive categories in regional zone. The research involved 36 farms. From data obtained it is shown that the number of pigs is 4273, of which 526 sows (43.1%), 58 boars (1.36%), 1693 piglets younger than 4 months (39.6.%), 1394 pigs between 4 and 12 months (32.6%), 602 pigs older than one year (14%), and an estimated average of an annual production of 4573 pigs. On this basis, it seems interesting to observe how the Nebrodi Black Pig farming it’s becoming an economic reality in expansion. It is hoped that in future this may represent an added value to promote the area.

Keywords. Nebrodi black pig – Management – Livestock system.

Le Porc Noir des Nebrodi: Description de la situation régionale avec une attention particulière à l'environnement et les techniques et modes d'élevage.

Résumé. *Le porc noir, élevé sur les Nebrodi, est un héritage familial transmis par les générations précédentes. Bien que l'élevage des races modernes présente un important avantage économique, c’est grâce aux interventions du Département pour l’agriculture et les forêts en Sicile, qu’il a été possible d’éviter la perte et l’extinction de ce patrimoine génétique. Les caractéristiques organoleptiques distinctives de la viande et de la graisse en particulier, combinées avec l'assaisonnement typique, font que les produits soient particulièrement appréciés par les consommateurs. L'objectif de cette recherche, qui s'inscrit dans un contexte plus large, est d'évaluer la diffusion et la quantité d’exploitations agricoles dans les différents secteurs productifs de la région. L'enquête a porté sur 36 exploitations agricoles et il en résulte que les porcs sont au nombre de 4273, dont 526 truies (43,1%), 58 verrats (1,36%) 1693 porcelets de moins de 4 mois (39,6%), 1394 porcs entre 4 et 12 mois (32,6%), 602 porcs de plus d’une année (14%), et on estime une production moyenne annuelle de 4573 porcs. Sur cette base, il semble intéressant de noter que l'élevage des porcs noirs des Nebrodi va devenir une réalité économique en expansion. On espère qu'à l'avenir, cela puisse représenter une valeur ajoutée pour promouvoir le terroir.*

Mots-clés. *Porc Noir des Nebrodi – Gestion d'entreprise – Modes d'élevage.*

I – Introduction

The Black Pig breeded in the Nebrodi' s area represents a cultural heritage that has been trasmitted by previous generations. In the last decade, Sicilian Region's Agriculture and Forestry Department has been sensitive to issues related to breeding, processing and marketing of products that are obtained from this breed of pig. Already in 2005, it initiated a project to improve the breeding system, defense of livestock biodiversity as a factor for territory development. This particular interest has found its international expression in Qubic Animal Breeding project: Quality Biodiversity Innovation Competitiveness included in the 2007-2013 MED Programme. For nearly two decades, in order to obtain a major productivity and consequently a revenues higher, the pig farmers have used commercial pig breeds, and local breeds have been relegated to a breeding to more amateur type than economic. This situation, of course, has led to a reduction in the number of local breed. Today, the Black Pig is included in the list of breeds at risk of extinction. Besides, the Nebrodi's Black Pig is a slow food presidia that is particularly popular for both the processed products than fresh meat, for which since 2005 the procedure to have the PDO quality sign has been initiated.

According to data of ANAS, the Black Pig breeding in Sicily has a different distribution in the provinces of Enna, Messina, Palermo, Ragusa and Siracusa (Table 1).

Table 1. Distribution of black pig breeding in Sicily

District	Farms Number	Percentage
Enna	2	5.1 %
Messina	24	61.5 %
Palermo	6	15.4 %
Ragusa	3	7.7 %
Siracusa	4	10.3 %
Total	39	

In the province of Messina there are the greatest number of officially recorded breedings (24 - 61.5%), but it is estimated that in territory they are present over a hundred. The spread of these breedings has its larger place in the territory of the Nebrodi' s Regional Park. The Nebrodi' s Black Pig find the best conditions for adaptability in the orographic and climate features of this region.

The aim of the survey, which is part of a broader study, is to provide informations on the area where the Nebrodi's Black Pig is bred mainly, on its distribution and abundance of animals in the involved farms.

II – Materials and methods

The survey involves 36 farms of Nebrodi's Black Pig located in municipalities of Sinagra, Mirto, Militello Rosmarino, S.Fratello, Longi, Frazzanò, Ucria, Castell'Umberto, Tortorici, Alcara Li Fusi, Naso, Caronia, Floresta, Capizzi and S. Lucia del Mela. A large part of them are located within the protected area of the Nebrodi Park (Fig.1).

The data collection of the farms of the sample involved was obtained through an interview to the Nebrodi black pig farmers.

The structured interview consists of the following areas of investigation:

- data about the environment (orographic description, type of vegetation zone elevations);
- data about management system;
- data on on the number of animals on farms.

Fig. 1 Georeference of farms and area of Nebrodi Park.

III – Results

1. Environment

Nebrodi's Regional Park, established in 1993, with its 86,000 ha is the largest protected natural area of Sicily. The Nebrodi, Madonie and Peloritani mointains constitute the Sicilian Apennines. They border on the north by the Tyrrhenian Sea, while its southern boundary is marked by Etna mountain, in particular from the Alcantara and Simeto rivers. Altimetric profile changes from few meters to 1847 meters (Soro mountain). Other mountais are Serra del Re (1754 m), Pizzo Fau (1686 m) and Serra Pignataro (1661 m) The elements characterizing the Nebrodi's andscape are the asymmetry of each slope, the different shaping of the mountains, the lush vegetation and wetlands and the presence of extensive areas of clay rocks and sandstone. Where there are the limestones, the landscape takes on aspects Dolomites like, with irregular outlines. This is the case of San Fratello mountain, and especially the Rocche of Crasto (1315 m).

The presence of forest has a significant impact on the climate of the Nebrodi area, which is characterized by long and harsh winters, and warm summers but not sultry weather. Temperatures in the hinterland generally remain between 10 and 12°C in the middle and high mountains, while the rainfall has a range from a minimum of 600 mm to a maximum of 1400 mm. The Nebrodi flora is related to slope (Table 2).

Table 2. Nebrodi vegetation

Plain Coastline and hills behind, up to 700-800 meters above sea level band thermomediterranean	The vegetation is represented by forests of evergreen *Quercus suber* alternating areas of Mediterranean scrub, which includes heather, the gorse (*Calycotome spinosa*), strawberry tree (*Arbutus unedo*), myrtle (*Myrtus communis*), euphorbia (*Euphorbia dendroides*), mastic (*Pistacia lentiscus*) and oak (*Quercus ilex*).
Hill Vegetative strip above, up to an altitude of 1000-1200 m above sea level band mesomediterranean	Formation of deciduous forests of *Quercus gussonei*, and a type of oak, *Quercus* and congested in some areas, the oak (*Quercus ilex*). In areas not forested blackthorn (*Prunus spinosa*), hawthorn (*Crataegus monogyna*), *Rosa canina, Rosa sempervirens*, the wild apple (*Malus sylvestris*), *Pyrus* and *Rubus amygdaliformis ulmifolius*.
Mountain Beyond 1200 we enter the mountain area proper band supramediterranea.	Extensive woodlands of oak (*Quercus cerris*) and beech (*Fagus sylvatica*) and the presence of mountain maple (*Acer pseudoplatanus*). The luxuriant underwood has a number of plant species among which are the holly (*Ilex aquifolium*), butcher's broom (*Ruscus aculeatus*), hawthorn (*Crataegus monogyna*) and yew (*Taxus baccata*).

1. Management system

The pasture management is structured as following (Table 3, A): 10 farms use a permanent agronomic system (38%), while 16 farms use a turnover system (62%). In the farms, where is present forest, it is an important souce of nutrition. In none of farms is observed soil erosion.

In farms with forest area (Table 3, B), 19 of them (58%) use to let the pigs in forest during all year, while 14 of them (42%) use to let the pigs in forest only in autumn-winter period or spring-summer, depending from climate conditions.

Table 3. Agronomic management system of pastures and permanence in forest areas

A	Pasture agronomic system permanently	38 %
	Pasture agronomic system turnover	62 %
B	Season	42 %
	All year	58 %

2. Number of animals in farms

The farms are divides by number of sows (Fig. 3 A), boars (Fig. 3 B) and production annual average (Fig. 3 C).

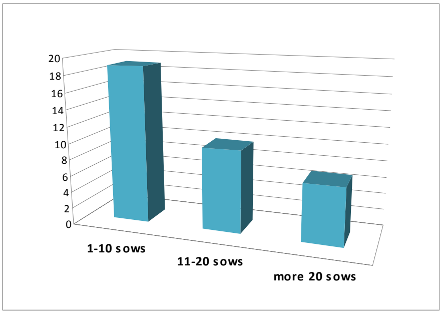

Fig. 3 A. Farm sizes expressed as the number of sows per class.

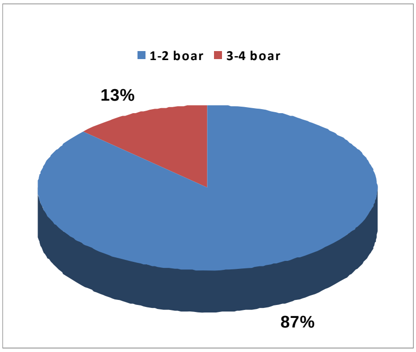

Fig. 3 B. Number of boar in farm.

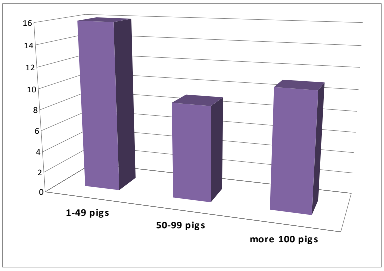

Fig. 3 C. Breeding size annual average.

As shown in Fig. 3, 83.3% of farms have fewer than 20 sows, while 87% have 1 or 2 boars. The average number of sows by farm is 14.6. Most farms have a production about 50 pigs by year.

At the time of the survey, in the farms there were about 4573 pigs of different commercial category and classified by class number of sows.

Table 4. Average annual production

	Number of sows			Total
	0 – 10 (M ± SD)	**11 – 20 (M ± SD)**	**> 20 (M ± SD)**	
Pigs < 4 months	23,2 ± 11,5	47,5 ± 23,3	111 ± 48,5	1693
Pigs between 4 and 12 months	17,4 ± 11,3	34 ± 21,4	103,5 ±61,3	1394
Pigs > 12 months	3,5 ± 7,7	24 ± 24,1	42,1 ± 60,1	602
N° of boars	1,1 ± 1,04	1,7 ± 0,64	2,85 ± 0,84	58
Sows / Boars	4,52 ± 1,76	9,85 ± 3,43	14,98 ± 6,15	526/58=9,06
Estimated average annual production	44,36 ±15,1	131 ±65,8	345,7 ± 169,2	4573

The average annual production by sow is 8.7 piglets. The average of annual production by farm is 127 pigs. The rate sows/boars by farm is 9.1. The 89% of the farmers do not make crossbred with other races.

IV –Conclusions

Thanks to the interest of the Sicilian Region’s Agriculture and Forestry Department and to the initiative carried out to the protection of this local breed, it is interesting to note that this data suggest a productive realty rather satisfactory. It is certainly a point of starting for a real push towards an increase in the chain of production and marketing of excellence local products obetained from this autochthonous breed.

The survey, in fact, shows a growing awareness of the farmer towards genetic resources, as well as data highlight:

89% of farmers are aware that it is extremely important to keep the animals in purity not crossbreding them with other races. This is certainly in favor of "quality". On the other hand, the road is still long in order to recognize fully the products derived by this animals. It is necessary, however, a deeper understanding of reality and of the territorial and economic management in order to define the parameters most relevant to the achievement of high quality products. Proper visibility of these products also means protecting and ensuring food security that consumer requires increasingly persistence.

Acknowledgements

The authors wish to acknowledge the financial support received from the Programme MED / MED Programme 2007-2013 / QUBIC Animal Breeding: Quality Biodiversity Innovation Competitiveness, Sicily Region - Department of Agriculture.

Want to thanks for technical assistance to: E. Rappazzo, G. Rubino, G. Romeo, A. Lastra, S. Bellinvia.

Effect of litter size on post-weaning growth of Iberian piglets

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Abstract. The current Iberian pig breeding program genetically evaluates sires and dams for 30-day and 90-day weight traits. However, very few studies relating the effect of litter size on piglet weight and growth have been published for this breed. Therefore, this study evaluates the effect of litter size on piglet weight and growth. Data consisted of 6,300 individual weights of piglets from the data-base of the Iberian Pig Association (AECERIBER) and belonging to 4 commercial herds. Weights were standardized to 30, 60, 90 and 120 days, and pre-and post-weaning average daily gains were calculated. The General Linear Model Procedures of SAS were used for data analysis, including the following fixed effects: herd and farrowing season; piglet sex; and litter size at birth, at 24 hours and at weaning. Piglet growth patterns from birth to 120 days were described. Litter size significantly affected these weights, although the effect of the increase in litter size was not constant, except in the case of birth weight. Perinatal mortality was greater in large litter sizes.

Keywords. Iberian piglets – Post-weaning growth – Litter size – Mortality.

Effet de la taille de la portée sur la croissance post-sevrage des porcelets Ibériques

Résumé. *Le schéma actuel de sélection pour la race de porc Ibérique évalue génétiquement les reproducteurs pour les caractères de poids à 60 et 90 jours, mais il n'y a pas beaucoup d'études sur l'effet qu'a la taille de la portée sur la croissance chez les porcelets vivants. Ce travail évalue l'influence de cet effet sur les poids et les croissances post-sevrage des porcelets. On a utilisé 6.300 mesures individuelles de porcelets originaires du schéma de sélection ACERIBER, qui appartenaient à 4 élevages. Les poids ont été classés à 30, 60, 90 et 120 jours et on a calculé les gains de poids pré et post-sevrage. Pour l'analyse on a utilisé la procédure des modèles linéaires SAS, incluant les effets fixes suivants : élevage et époque de la mise bas ; sexe du porcelet ; et taille de la portée à la naissance, à 24 heures et au sevrage. On décrit les tendances de croissance des porcelets depuis leur naissance jusqu'à 120 jours. La taille de la portée a affecté significativement les poids mentionnés, bien que l'effet de l'augmentation de la taille de la portée n'ait pas été constant, sauf pour le poids à la naissance. La mortalité périnatale a augmenté avec la taille de la portée.*

Mots-clés. *Porcelets Ibériques – Croissance – Post-sevrage – Taille de la portée – Mortalité.*

I – Introduction

Litter size and pre- and post-weaning growth are very important factors for the profitability of swine production. In the Iberian pig, for which mean litter size usually do not surpass 7 piglets (Suárez *et al*., 2002), it would be of great value to know how litter size affects the subsequent piglet growth. With this knowledge, producers would be able to determine the maximum liter size that allows for a greater yield without significantly decreasing piglet growth. Most studies on Iberian pig production in this area analyzed pre- and post-weaning traits regardless litter size. However, several studies have been done in white swine breeds determining the effect of litter size on individual piglet growth during lactation (Canario *et al*., 1993). Therefore, litter size could be increased through genetic improvement (Bidanel *et al*., 1994; Johnson *et al*., 1999; Noguera *et al*., 2002), aided by adequate management procedures, up to an optimal degree for maximal profitability. In consequence, the objective of this study was to determine the effect of litter size traits on the post-weaning growth of Iberian piglets.

II – Materials and methods

The present study analyzed approximately 6,300 individual weighings from purebred Iberian piglets, registered in the breeding selection program of AECERIBER (Spanish Association of Iberian Pig Breeeders) and belonging to four enterprises and to the Valdesequera research farm of the Agricultural Research Center of Extremadura (SW Spain). The management system for the farrowing and pre- and post-weaning phases was the same for all the farms involved in the study. Data obtained from the farrowing and lactation phases were: total number of piglets born (TB); number of piglets born alive (BA, which are calculated as TB minus those born dead or those died within 24 hours post-partum); and number of piglets weaned (W). The average weaning age was 30 days. Individual weighings were performed for each piglet within each litter at birth and between 21 and 140 days of age, and these body weights (BW) were standardized to 30, 60, 90 and 120 days. Data were analyzed with the SAS statistical package using a linear model in which BW were corrected for "farm", "sow age at farrowing", "season", and "piglet sex". For each level of litter size, measured as numbers of TB, BA and W, the following variables were obtained: corrected mean individual piglet weight at birth; and mean individual BW standardized at 30, 60, 90 and 120 days, which were the ages having more BW measurements. The effect of litter size (TB, BA, W) on weight at birth, at 30 days (similar to weaning weight) and at 60, 90 and 120 days of age was determined.

III – Results and discussion

Results for birth weight are depicted in Fig. 1.

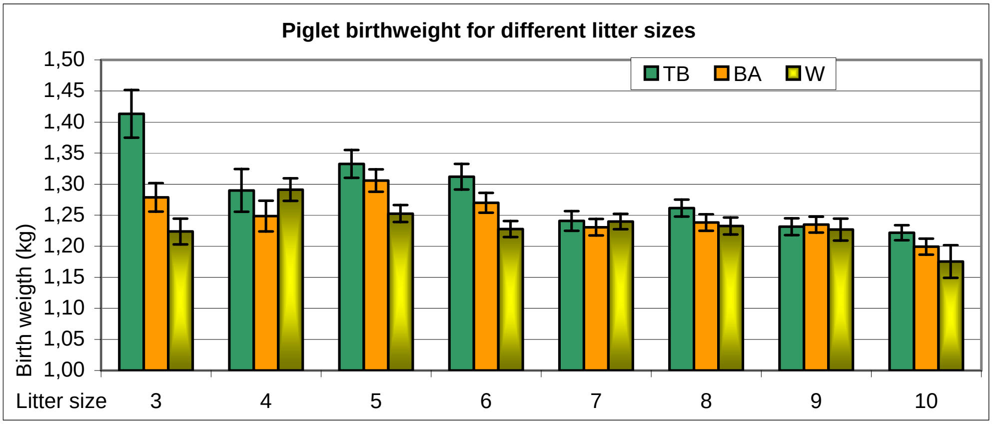

Fig. 1. Effect of litter size, in terms of total born (TB), born alive (BA) or weaned (W) piglet number, on piglet birth weight.

Litters with 3 TB had superior birth weight than litters with more than 6 TB piglets. Also, litters with less than 7 piglets were more heterogeneous than litters with 7 to 10 piglets, as indicated by the greater coefficients of variability of TB classes 1 to 6 (data not shown). Therefore, increasing litter size up to 10 piglets may not affect litter homogeneity. Litters with 3 or 4 BA piglets had birth weights significantly smaller than litters having 5 BA piglets. Birth weight decreased gradually and significantly from 5 BA to 7 BA piglet litters, but it did not significantly change from 7 to 9 BA-piglet litters. However, it significantly decreased from 9 to 10 BA-piglet litters. Analogously to the above mentioned TB coefficients of variability, litters having 7 to 10 BA piglets were more homogeneous at birth than those containing less BA piglets (data not shown). In contrast, birth weight did not significantly change as the number of W piglets increased, except for the 10 W piglet class, which had a significantly smaller birth weight than the other classes, and also the 4 W piglet class, which had greater birth weight than the rest. Other authors, like Kaufmann *et al*. (2007), found greater differences in birth weight as the number of weaned piglets increased.

Figures 2, 3 and 4 depict the effect of litter size (in terms of TB, BA or W piglet numbers, respectively) on piglet BW at 30 (BW30), 60 (BW60), 90 (BW90) and 120 (BW120) days of age. Figure 2 shows no effect of TB number for BW30, BW60 or BW90, except for the 14 TB class, which corresponded to smaller BW values, although there were few litters belonging to this class. Nevertheless, litters having 11 or 12 TB piglets had smaller BW120 values than the remaining litters.

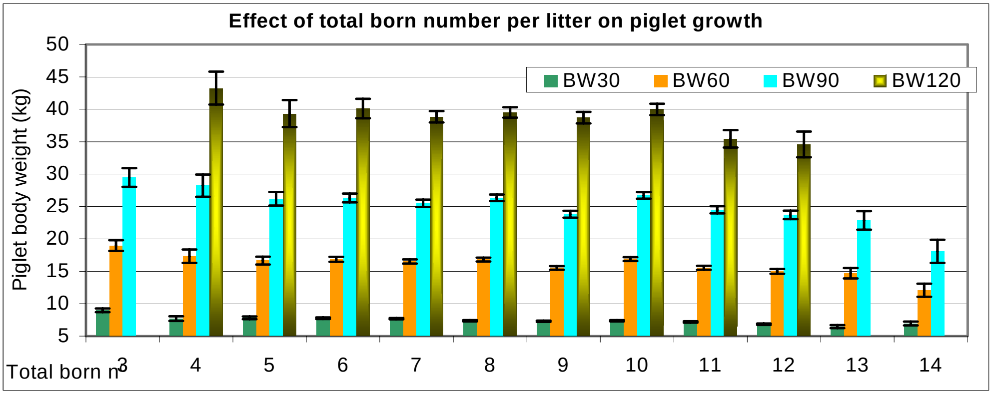

Fig. 2. Effect of number of total born per litter on piglet body weight at 30 (BW30), 60 (BW60), 90 (BW90) and 120 (BW120) days of age.

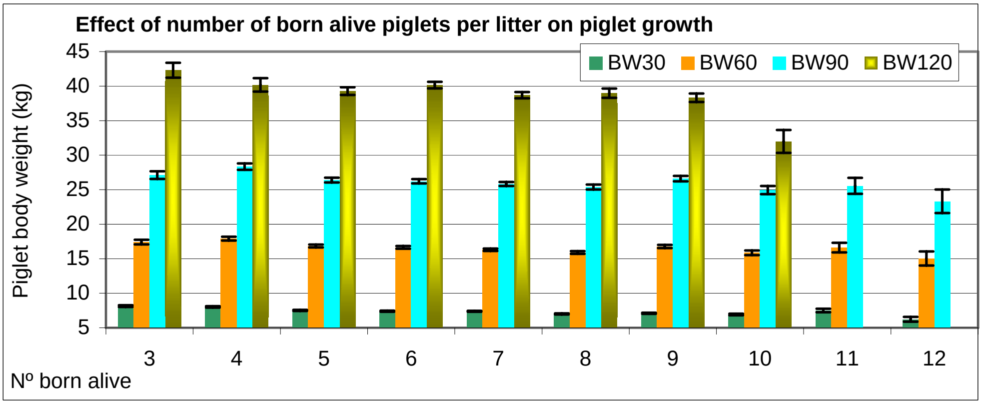

Fig. 3. Effect of number of piglets born alive per litter on piglet body weight at 30 (BW30), 60 (BW60), 90 (BW90) and 120 (BW120) days of age.

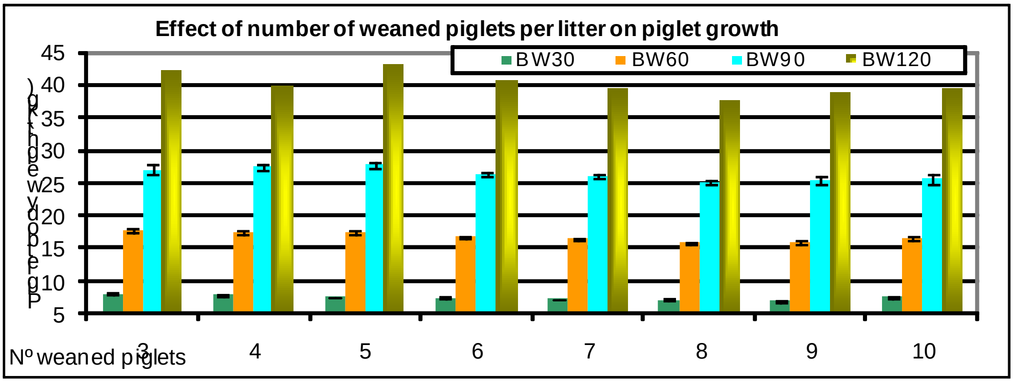

Fig. 4. Effect of number of weaned piglets per litter on piglet body weight at 30 (BW30), 60 (BW60), 90 (BW90) and 120 (BW120) days of age.

Figure 3 represents piglet BW progression depending on BA piglet number. Piglet BW30 was greater in litters containing 3, 4 or 5 BA piglets, decreased from the BA 6 to the BA 11 classes, and was smaller for the BA 12 class. The number of BA piglets did not affect piglet BW60 nor BW90, except for the 12 BA class, for which both weights were smaller. Finally, BW120 was greater when BA number was 3, was constant from the 4 to 9 BA classes, and significantly decreased for BA numbers greater than 9.

Figure 4 depicts the differences in piglet BW at different ages depending on the number of weaned piglets. When the number of weaned piglets was greater than 5, all BW values decreased somewhat, reaching the significance level for BW30, BW90 and BW120.

In conclusion, a possible decrease in piglet weight can be expected as the number of weaned piglets increases, but this decrease should not be detrimental from the economical point of view because it is compensated by the greater number of piglets weaned per litter.

Acknowledgements

The authors want to thank FEDER, (PDT09B039), INIA (RTA2007-0093-00-00) and Junta de Extremadura for financial support.

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Study on the behaviour of Cinta Senese sows and piglets reared outdoors

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Abstract. The aim of this study was to observe the behaviour of Cinta Senese sows and their litters reared in outdoor systems. Data were collected on 4 sows weekly for 24 consecutive hours (1-hour intervals) and submitted to analysis of variance with fixed effects: season (spring and summer) and day slots (morning, afternoon, evening and night). The behavioural models were collected into 5 main activities for sows and 6 for piglets. Considering only the main effects, sows spent the most time at rest, especially in summer (P<0.01), and in the afternoon (77 %) and night (76 %) and therefore the search for food occurred mainly in morning (15 %) and evening (37 %). The activity of rooting (spring > summer; P<0.01) occurred also mainly in morning and evening (30 and 15 %, respectively). In addition, piglets spent much time at rest; in particular, during the first weeks of life they preferred to stay in group (44 %) rather than isolates (13 %). Time spent for feeding was highest in spring and in morning and evening (P<0.01). Rooting activity was influenced by season and day slot (P<0.01).

Keywords. Sow behaviour – Piglets behaviour – Outdoors rearing – Animal welfare.

Observations sur le comportement des truies et des porcelets Cinta Senese en élevage extensif

Résumé. *L'objectif de cette étude était d'observer le comportement des truies Cinta Senese et leurs portées élevées en libre parcours. Les données de 4 truies, recueillies chaque semaine, pendant 24 heures consécutives (intervalles de 1heure) ont été soumises à l'analyse de variance avec des effets fixes : la saison (printemps et été) et le créneau horaire (matin, après-midi, soir et nuit). Les modèles de comportement ont été regroupés en 5 activités principales pour les truies et 6 pour les porcelets. Les truies passent le plus de temps au repos, surtout en été (P<0,01), en favorisant l'après-midi (77%) et la nuit (76%) et donc la recherche de nourriture a eu lieu principalement dans la matinée (15%) et la soirée (37%). L'activité de fouissage, (printemps > été ; P<0,01) s'est produite principalement pendant le matin et le soir (30 et 15 %, respectivement). Les porcelets passent également beaucoup du temps à se reposer, et en particulier pendant les premières semaines de vie ils sont davantage restés en groupe (44%) qu'isolés (13%). Le temps consacré à l'alimentation était le plus élevé au printemps et au cours du matin et du soir (P<0,01). L'activité de fouissage a été influencée par la saison et l'heure de la journée (P<0,01).*

Mots-clés. *Comportement des truies – Comportement des porcelets – Élevage extensif – Bien-être animal.*

I – Introduction

In intensive farming systems, pigs are often kept in unsuitable environments to meet their behavioural needs, that are important indicators of "welfare" (Fraser *et al*., 2001). The complexity of breeding may influence the cognitive ability of pigs (De Jong *et al*., 2000; Sneddon *et al*., 2000) and their ability to cope with stress (De Jonge *et al*., 1996; O'Connell *et al*., 1999). The outdoor system offers more space for pigs, a more stimulating environment and free access to food compared with the intensive system. This may allow the animals to implement a rich repertoire of behaviour and thus develop the skills necessary to cope with stress. The amount of social contact with the sow during the lactation period, which differs between the two systems (Cox and Cooper, 2001) can be determined by the commitment of piglets in beneficial activities for their adaptation to weaning, how to eat solid food, explore the environment and interact with other broods. The aim of this work was to study the behaviour of sows and piglets of Cinta Senese breed reared outdoors. The results related to feeding, static and dynamic behaviour, observed in two different seasons: spring and summer.

II – Materials and methods

The surveys were carried out on 4 sows and their litters, with a total of 29 piglets in the period from birth to weaning (78 d), two sows gave birth in spring and two in summer. Data were collected weekly noting the behaviour each hour for 24 hours. The day was divided into 4 day slots: morning (6-11 h), afternoon (12-16), evening (17-20) and night (21-5).

The semi-outdoor rearing system consisted of shelters with paddocks and fences for grazing that was round. The grazing rhythms were adapted to the seasonal photoperiod and at the return in the shelters was given supplementation with concentrates.

The different types of observed behaviours have been grouped into five main activities (Table 1).

Table 1. Definition of the specific behaviours that were observed

Main activities	Sow	Piglet
Resting	The sow is laying or sitting or sleeping.	Alone: the piglet is laying alone or with his mother; sitting or sleeping. In group: the piglet are laying in group closed other piglets; sitting or sleeping.
Eating	The sow has its head inside the feeder or the sow is eating straw or grass.	Piglet has its head inside the feeder or piglet is eating straw or grass.
Moving	The sow is standing or walking, or touching another pig in some way or is doing other social behaviours (nursing or grooming).	The piglet is standing or walking, or touching another pig in some way or is doing other social behaviours (grooming).
Rooting	The sow is rooting or trying to root on the floor/in the mud or in the deep straw.	Piglet is rooting or trying to root on the floor/in the mud or in the deep straw.
Suckling	The sow allow the piglets to massage or suck at the udder; the sow is emitting vocalizations.	Piglet massages or sucks at the udder of their mother or of another sow, or piglet researches udder; piglet is emitting vocalizations.

The relative frequencies of the main activities, compared to zero (absent), were subjected to analysis of variance, considering as fixed effects the season and the day slot and their interaction, through the proc. GLM of SAS statistical package (SAS Institute Inc., 2003).

III – Results

Behaviour of the sow: Table 2 reports the average percentages collected for the different sow's activities.

The rest was influenced by season and day slot. Sows spent more time resting in summer and spring, focusing on the afternoon and night in both seasons (data not tabulated), in partial agreement with De Passillé and Robert (1989) which found that sows in cages remained lying more during the hours of darkness than in the hours of light.

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Table 2. Effect of season and day slot on the sow's behaviour (%)

Activities	Season		Day slot				
	Spring	Summer	Morning	Afternoon	Evening	Night	DSR
Resting	43.81 b	53.75 a	26.66 a	77 b	15.63 c	75.83 b	20.32
Alimentation	14.33	16.77	15 a	2 b	36.87 c	8.33 b	17.32
Rooting	19.70 a	5.03 b	29.58 a	3.50 b	15 c	1.39 b	13.67
Moving	6.98 b	11.60 a	16.25 a	5.50 b	13.75 a	1.67 b	12.91
Suckling	15.18	12.83	12.50	12	18.75	12.78	15.11

Within criterion, means with different letters differ ($p<0.05$)

The eating activity occurred differently in the day slots that interacted with the season. Indeed, in respect to spring, in summer sows are more active in seeking food in the morning and vice versa in the evening (Fig. 1). However, the more eating activity during summer is explained by the fact that the animals were given greater permanence to pasture. The rooting also is influenced by both factors that significantly interacted themselves. The sows spend more time rooting in the morning and evening, especially in spring; instead in summer the sows devoted few time to this activity (Fig. 2). This could be due also to different quality of available grazing in the seasons, which has stimulated animals to research food. The suckling activity is not influenced by external factors.

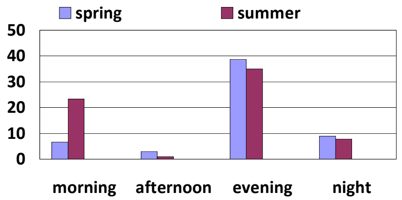

Fig. 1. Eating activities of sows for season and day slot (% of time).

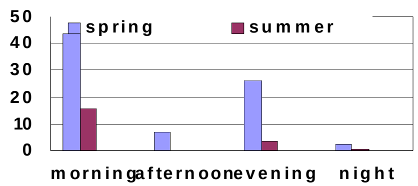

Fig. 2. Rooting activities of sows for season and day slot (% of time).

Behaviour of piglets: Table 3 shows the average percentages reported for the various activities of the piglets. When alone, they rested, more in the summer than in spring and especially during the afternoon (interaction significant at $p<0.01$). In group, on the contrary, they rested more in the afternoon or at night.

Considering together the results for the resting, in spring piglets rested almost always in groups, while in summer the alone resting has increased in each day slot, (Figs 3 and 4).

Eating activity of piglets was influenced by season, day slot and the interaction between the factors and they devoted more time to feeding in spring than in summer and in both seasons in the evening hours (Fig. 5).

Table 3. Effect of season and day slots on the piglet's behaviour (%)

Activities	Season		Day slot				
	Spring	Summer	Morning	Afternoon	Evening	Night	DSR
Resting in group	46.97	40.93	39.04 b	50.50 b	22.36 c	63.91 a	28.38
Resting alone	4.61 a	21.64 b	4.01 b	22.34 a	9.89 b	16.27	22.32
Alimentation	10.93 a	6.85 b	9.74 c	3.58 b	21.09 a	1.14 b	12.09
Rooting	6.71 a	3.48 b	12.26 a	2.37 c	5.75 b	0 c	7.17
Moving	11.19	11.53	20.27 a	6.28 b	16.77 a	2.11 b	11.34
Suckling	18.29	15.50	14.79	14.80	21.69	16.31	15.63

Within criterion, means with different letters differ (p<0.05).

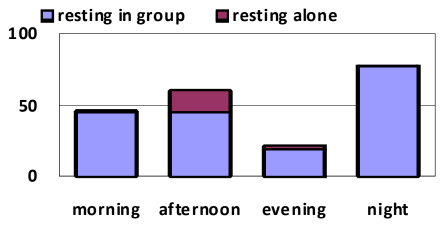

Fig. 3. Resting activities of piglets in spring (% of time).

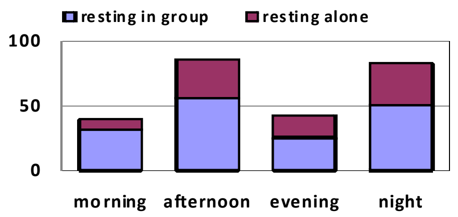

Fig. 4. Resting activities of piglets in summer (% of time).

Also the activity of rooting was influenced by season and day slot, with greater activity in spring (Fig. 6). Be noted that the food management technique adopted favoured rooting in the morning (grazing animals) and eating in the evening (feeding).

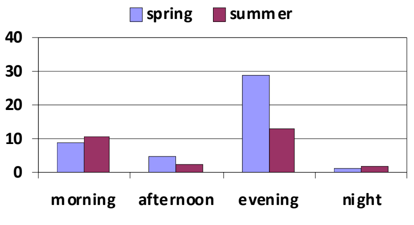

Fig. 5. Eating activity of piglets for season and day slot (% of time).

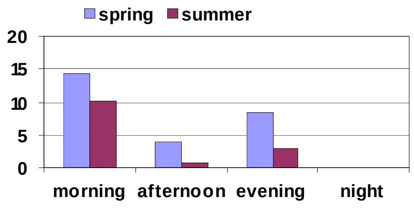

Fig. 6. Rooting activity of piglets for season and day slot (% of time).

IV – Conclusions

The observations show that pigs reared under extensive system have the opportunity to express for many time specific behaviours such as rooting, which is not possible in an intensive farming. This technique of animal management, with the consequent movements, influenced mainly food-related activities.

Almost all activities are influenced by season and day slot, confirming the strong conditioning of the environment on animals in the extensive system.

The suckling activity finally, was the least affected by environmental factors.

Acknowledgements

The Authors want to thank mrs dr. Maria Novella Uzielli of “S.p.A. Tenuta di Paganico” farm. The research was supported by ARSIA Toscana

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Study of fertility, fetal survival and prolificacy in relation to social rank in multiparous Iberian sows

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Abstract. The causes of the low prolificacy of the Iberian pig are unknown, and embryo mortality has not been characterized in this breed, for which breeding and gestation are usually managed in groups, thus resulting in stress related to social hierarchy that could affect its reproductive efficiency. Therefore, the objective of this study was to determine the influence of social rank on fertility, embryo mortality and prolificacy in the Iberian pig. Multiparous Iberian sows (n=39) were weaned at 35±3 postpartum days and then were bred in 4 corrals using 4 boars during ≈40 days. Social rank of sows (R1=dominant; R2=intermediate; R3=subordinate) was determined during feeding, using the 4 corrals up to ≈90 days post-weaning and then a single, larger corral. Sows were scanned by ultrasonography at 20-30, 35-45 and 50-60 post-coitum days. Body weight was similar among ranks. Conception rate at first estrus was lower for R3. Fetal survival after 40 days of gestation was greater for R2 than for R1 and R3. However, R3 sows tended to farrow more live piglets and to yield heavier piglets at weaning.

Keywords. Iberian pig – Behavior – Litter size – Embryo mortality – Ultrasonography.

Etude de la fertilité, survie embryonnaire et prolificité en fonction de la hiérarchie sociale chez les truies multipares de porc Ibérique

Résumé. *Les causes de la faible prolificité du porc Ibérique ne sont pas connues, et la mortalité embryonnaire n'a pas été caractérisée pour cette race, chez laquelle on a l'habitude de manipuler ensemble la mise à la reproduction et la gestation, résultant en un stress dérivé de la hiérarchie sociale qui pourrait diminuer son efficacité reproductive. L'objectif de cette étude était d'identifier l'influence du rang social sur la fertilité, la mortalité embryonnaire et la prolificité chez le porc Ibérique. Des truies Ibériques multipares (n=39) ont été sevrées à 35±3 jours post-partum, et elles ont été placées dans 4 enclos différents avec 4 verrats pendant 40 jours. Le rang des femelles (R1=dominantes; R2=intermédiaires; R3=subordonnées) a été déterminé pendant la distribution de la nourriture, en utilisant les 4 enclos jusqu'à ≈90 jours après le sevrage, et un enclos plus grand ensuite. Les truies ont été échographiées à 20-30, 35-45 et 50-60 jours post-coïtaux. Le poids corporel était similaire entre rangs. Le taux de conception à la première chaleur était inférieur pour R3. La survie fœtale postérieure à 40 jours de gestation était plus élevée pour R2 que pour R1 et R3. Cependant, les R3 tendirent à mettre bas plus de porcelets vivants et à les sevrer plus lourds.*

Mots-clés. *Porc ibérique – Conduite – Prolificité – Mortalité embryonnaire – Ecographie.*

I – Introduction

Many studies have been published reporting productive data related to reproduction in the Iberian pig, but the information about the reproductive physiology of the Iberian female is very scarce, probably due to the limited geographic distribution of this breeed. In fact, the ovulatory, uterine and fetal components related to litter size in this breed are still unknown. However, this information is really needed for this breed, which is known by its low prolificacy in comparison with the main European breeds (Dobao *et al*., 1988; Vázquez *et al*., 1994). In the Iberian pig, the mean litter size is seldom greater than 7, and it increases with parity up to a maximum mean of 9.5 from the 5th to the 8th farrowing (Vázquez *et al*., 1994). Besides, the new European regulations on animal welfare make group housing of female pigs during gestation mandatory, which poses many questions about the influence of stress on reproduction in this and other breeds. Stress occurring close to ovulation can delay or inhibit it in farm animals (Dobson and Smith, 1995), and a low welfare level may decrease prolificacy in pigs (Robert , 1992). In addition, the social rank of group-housed female pigs, which is attained by stressful, aggressive interactions, affects reproductive efficiency and results in lower fertility and prolificacy in lower-rank females (Hoy *et al*., 2009). Therefore, stress arising from hierarchical interactions among females must be considered in modern pig production, and more specially in the case of the Iberian pig, which being an old breed might be more sensitive to social rank related mechanisms for population regulation, as it is known to happen in many wild mammals. Moreover, the usually extensive management of Iberian females may exacerbate these hierarchical interactions. Thus, the objective of the present study was to determine the effect of social rank of group-housed Iberian sows on fertility, fetal survival and prolificacy.

II – Materials and methods

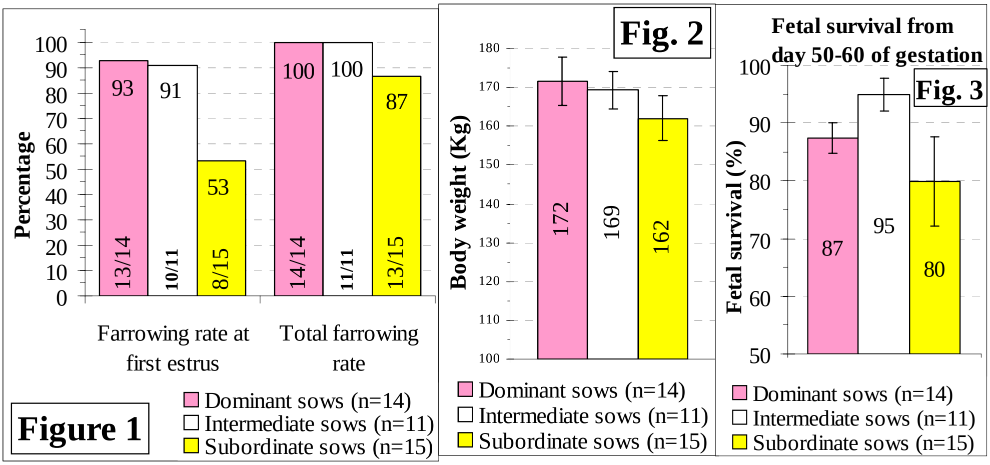
Multiparous Iberian sows (n = 39) were used in this study, which was conducted at the research farm of the agricultural Research Center of Extremadura (Southwestern Spain). Sows were weaned at 35±3 postpartum days (as usual for this breed) in 4 consecutive weaning lots, one lot every 7 days in June-July. After weaning, sows were housed in 4 outdoor corrals of ≈80x20 m with an Iberian boar in each. Systematic observations (8-10 AM and 5-6 PM) for estrous activity and estrous-related behavioral, hierarchical interactions were done during the first 8 days post-weaning. The mating period lasted for ≈40 days. Sows were weighed at the end of the mating period and remained in these 4 corrals for ≈50 more days after boar withdrawal. Then, at ≈90 days post-weaning, sows were joined in a large paddock until ≈7 days before farrowing, which took place in indoor farrowing crates, where the dams remained until the ensuing weaning. The social rank of the sows was evaluated by performing *food competition tests* (Andersen *et al*., 1999), which were performed in the 4 corrals, weekly during the mating period and every 2 weeks afterwards. Dorsal marking with paint spray aided to easily identify the individual animals. After boar withdrawal, and to improve rank evaluation, a greater range of rank scores was obtained by re-grouping the sows according to their previous rank. All the partial scores from each sow were used to obtain the overall rank: dominant (R1), intermediate (R2) or subordinate (R3).

Transabdominal ultrasound scanning was performed to diagnose pregnancy and to attempt the counting of conceptuses (embryonary vesicles or placental cavities of feto-placental units). Sows were scanned on 3 days, namely on post-coitum days 20-30, 35-45 and 50-60. A scanning crate, specially designed for this study, was used to restrain the animals while feeding, thus easing precise scanning and minimizing stress, which could alter the reproductive results. An Aloka-SD500 ultrasound scanner and 2 types of deep penetrating, 3.5 MHz ultrasound probes were used, one convex and narrow (4 cm) and another linear and long (17-cm transducer head). To count conceptuses, both uterine horns were scanned with the convex probe in the usual way, and then the long linear probe was used to check for accuracy of the spatial distribution of cavities. The digital images were stored and re-played in slow motion. Percentage continuous variables were arcsin-transformed before statistical analysis.

III – Results and discussion

The farrowing rate (Fig. 1) resulting from the first post-weaning estrus was smaller ($p<0.05$) for R3 sows (only 53%) than for R1 and R2. The total farrowing rate (including that resulting from repeating estruses) was 100% except for R3 sows, from which 1 did not conceive and another lost its pregnancy after day 58 post-coitum. These results suggest that the reproductive problems related to social rank and the possibly related stress occurred mainly for the lowest rank females, while the intermediate sows, interestingly, performed as well as the dominant animals. A possible explanation may be that R1 sows only could kept their attention on chasing the lowest rank females. It must also be noticed that social rank did not significantly depend on body weight ($p>0.1$; Fig. 2). This suggests that social hierarchy might be mainly based on psychological, behavioral factors related to aggressiveness instead of body strength related factors. Therefore, managerial decisions about female group arrangements should not be based only on body size.

Counting conceptuses was very difficult between 20 and 40 days of gestation because of the contorted placental cavities found. However, with the methodology applied, it was feasible to count them during the 3rd scanning (day 50-60 of gestation) in a repeatable way, because of their more regular shapes. Nevertheless, a slight overestimation was difficult to avoid because of the tight, crowded arrangement of large placentae at this stage, which frequently made fetuses and cavities show up twice. The presence of mummies further hampered image interpretation due to their blurry placental content. However, the overestimation of conceptus count (excluding a 12-fetus pregnancy lost) *vs* total born number (born alive + stillborn + mummies) vas not significant (10.3±0.4 *vs* 9.7±0.4; $p>0.1$), and their correlation was acceptably high ($r=0.74$; $R^2=0.54$; data not shown). Pig conceptuses can be reabsorbed only until day 35-40 of gestation, but most of the mummies formed during early pregnancy (before day 50) are too small to be detected at farrowing (Van der Lende & Van Rens, 2003). The fetal survival rate (Fig. 3) from the 3rd scanning with respect to the number of piglets born (either alive or stillborn, thus excluding the mummified fetuses) was higher ($p<0.05$) for R2 than for R1 and R3.



Figs 1-3. Fertility, body weight and fetal survival depending on social rank.

Moreover, R1 and R3 had 5 and 2 litters with mummies, respectively (13 mummies in total), and R3 totally lost a 12-fetus pregnancy after that scanning, but no mummies were found in R2 litters. Only these data, regardless the echographic study above described, yields also a greater fetal survival rate for R2 vs R1 and R3 (100±0% *vs* 94±2% and 90±7%, respectively; $p<0.05$; data not shown). This greater fetal survival of R2, again, suggests that R2 females had less stress, maybe because the R1 and R3 groups were more involved in aggressive or defensive behaviors. In contrast, the number of conceptuses present at the 3rd scanning was similar ($p>0.1$) for the 3 ranks (Fig. 4), which is consistent with other studies (Van Wettere *et al.*, 2008) showing no effect of hierarchical fighting related to re-grouping during early gestation on embryo survival.

The number of born alive + stillborn was similar among ranks (R1: 9.1±0.6; R2: 9.9±0.1; R3: 9.3±0.6; p>0.1,data not shown). However, R3 sows tended to have more born alive and less stillborn piglets (p<0.1; Fig. 4). Perhaps lower rank females are less stressable during parturition and, thus, have a lower incidence of hypoxic stillborn piglets. This good maternal trait could be consistent with less aggressive females, perhaps partially explaining the origin of their rank. Piglet weight at weaning tended (p<0.1) to be smaller for R3 *vs* R2 litters (Fig. 5). Coefficients of within-litter variation for growth traits were very similar among ranks (Fig. 5). At weaning, piglet survival and litter weight were also similar among ranks (Figs 4 and 6; p>0.1).

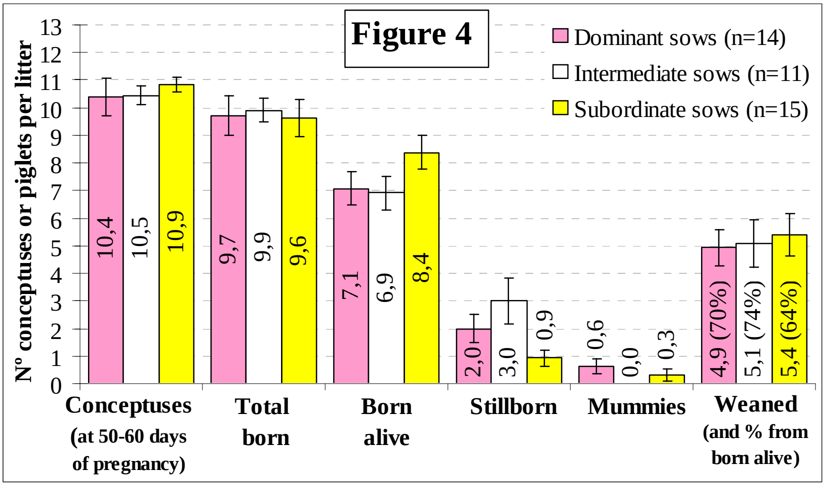
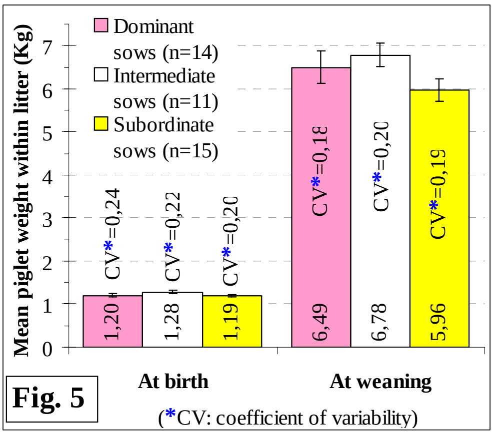
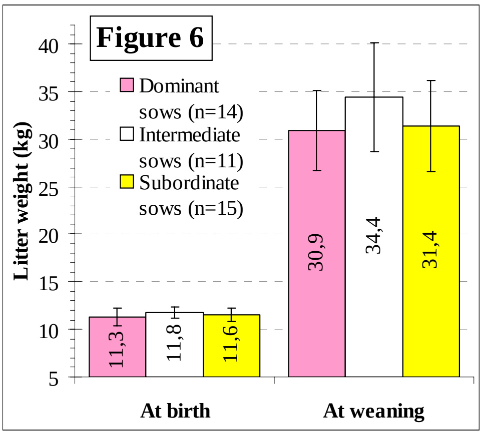


Fig. 4. Litter size. Due to conception failure and pregnancy loss, n=14-13 for Subordinate sows.





Figs 5-6. Piglet and litter weight at birth and weaning. Subord.: n=13 due to missed farrowings.

Having greater litters, this may imply that R3 females had good maternal traits. In conclusion, social rank affected reproductive traits mainly in R3 sows, as expected, but, surprisingly, R2 females were less affected than the dominant, R1 sows. In contrast, R3 sows may have superior maternal traits. Finally, social rank is not only based in body weight, which may be an issue to be considered when re-grouping females.

Acknowledgements

This study vas financed by *Junta de Extremadura* (3PR05A070) and FEDER (PDT09B039).

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Volatile fatty acids and meat composition

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Abstract. The digestion of fiber in the diet of pigs is mainly the large intestine. Volatile fatty acids (mainly acetic, propionic and butyric acids) are produced as a result of fermentation and are used metabolically after being absorbed through the intestinal wall. Linear odd-chain fatty acids (C15: 0 and C17: 0) can be synthesized de novo from propionate in adipose tissue. The aim of this study was to determine whether the presence of odd-chain fatty acids in the subcutaneous tissue is influenced by the production of propionic because fiber fermentation in the large intestine. A total of 20 Iberian pigs divided into five lots was used. The sugar beet pulp and cereal straw were incorporated as the major fiber sources in diets with levels of incorporation of 300, 450 and 600 $g{\cdot}kg^{-1}$. The subcutaneous fatty acid composition was determined at beginning of period (by biopsy) and at slaughtered. The FA composition varies depending on the type of diet given to pigs. Volatile fatty acids production does not change the composition of the odd-chain fatty acids.

Keywords. Volatile fatty acids – Fiber – Subcutaneous adipose tissue.

Acides gras volatils et composition de la viande

Résumé. *La digestion des fibres dans l'alimentation des porcs se fait surtout dans le gros intestin. Les acides gras volatils (principalement les acides acétique, propionique et butyrique) sont produits à la suite de la fermentation et sont utilisés après avoir été métaboliquement absorbés par la paroi intestinale. Les acides gras linéaires à chaîne impaire (C15:0 et C17:0) peuvent être synthétisés de novo à partir du propionate dans le tissu adipeux. L'objectif de cette étude était de déterminer si la présence d'acides gras à chaîne impaire dans le tissu sous-cutané est influencée par la production d'acide propionique, en raison de la fermentation des fibres dans le gros intestin. Un total de 20 porcs Ibériques divisés en cinq lots ont été utilisés. La pulpe de betterave sucrière et la paille de céréales ont été intégrées en tant que sources majeures de fibres dans l'alimentation selon des niveaux d'incorporation de 300, 450 et 600* $g \bullet kg^{-1}$. *La composition sous-cutanée en acides gras a été déterminée en début de période (par biopsie) et à l'abattage. La composition en AG varie selon le type d'alimentation donnée aux porcs. La production d'acides gras volatils ne modifie pas la composition en acides gras à chaîne impaire.*

Mots-clés. *Acides gras volatils – Fibres – Tissu adipeux sous-cutané.*

I – Introduction

The digestion of fiber in the diet of pigs is mainly the large intestine. The rate of degradation of dietary fiber depends on dietary fiber characteristics such as the chemical composition, the degree of lignification, particle size, or the water solubility. Volatile fatty acids (mainly acetic, propionic and butyric acids) are produced as a result of fermentation and are used metabolically after being absorbed through the intestinal wall. Propionate is involved in the synthesis of linear odd-chain fatty acids (C15:0 and C17:0) by two metabolic pathways: (A) by lengthening of the carbon chain by micro-organisms (Fulco, 1983; Kaneda, 1991), followed by absorption and uptake of these exogenous and circulating fatty acids by the tissues, (B) by fatty acid synthesis and incorporation of propionyl-CoA instead of acetyl- CoA during the synthesis of these endogenous fatty acids in milk secretion (Loor *et al*., 2005; Vlaeminck *et al*., 2004) and in adipose tissue (Berthelot *et al*., 2001). Whereas, regression analysis suggested cis-9 C17:1 to be a desaturation product of C17:0 (Fievez *et al*., 2003; Kay *et al*., 2005). However, all these studies have been conducted on ruminants (Berthelot *et al*., 2001) or rabbits (Papadomichelakis *et al*, 2010) but not in pigs.

The aim of this study was to determine whether the presence of odd-chain fatty acids in the subcutaneous tissue is influenced by the production of propionic acid because fiber fermentation in the large intestine.

II – Materials and methods

A total of 20 Iberian pigs divided into five lots was used. Sugar beet pulp SBP (as soluble fiber) and cereal straw CS (as insoluble fiber) commercially available were incorporated as the major fiber sources in diets according to the scheme presented in the Table 1. The diets were milled to pass through a 2 mm screen diameter and offered as granule. The chemical composition of the diets is presented in Table 1. The diets contained similar crude protein contents and total amino acids, calcium and phosphorus levels and were balanced for metabolizable energy (ME). The pigs started the experiment with an average live weight of 84 kg and remained there for 28 days, up to 102 kg of live weight and were then slaughtered.

Table 1. Content of fiber sources on the experimental diets ($g \cdot kg^{-1}$) and chemical composition ($g \cdot kg^{-1}$ dry matter)

Diets	1	2	3	4	5
Sugar beet pulp	150	300	150	225	300
Cereal straw	150	150	300	225	300
Crude fat	23.5	20.7	38.0	27.7	49.5
∑Saturated	7.6	6.2	12.6	8.8	18.6
∑Monounsaturated	7.7	5.9	15.7	9.6	22.8
∑Poliunsatured	8.0	8.5	9.4	9.1	7.7
∑Odd-chain fatty acid	0.2	0.1	0.3	0.2	0.4

It were taken samples from subcutaneous fat at beginning of period (by biopsy) and at slaughtered. The fatty acid (FA) composition of the lipids were determined by gas chromatography after acidic-trans-esterification in the presence of sulfuric acid (5% sulfuric acid in methanol) (Cava *et al*., 1997). The gas chromatograph, model Hewlett-Packard 4890 Series II, was equipped with a split/splitless injector and a flame ionization detector. FAs were separated on a nitroterephthalic acid modified polyethylene glycol semicapillary fused silica column (30 m long, 0.53 mm i.d., 1 μm film thickness).

To determine volatile fatty acids (VFA) concentration (acetic, propionic, butyric, isobutyric, isovaleric and valeric acid), samples from the caecum and proximal colon were centrifuged. The supernatant fraction was analyzed chromatographically for VFA essentially by the method of Brighenti (1997) (using 2-ethyl butyrate as an internal standard, rather than isovaleric acid).

Statistical analyses were performed using SPSS (version 15.0, 2008). One-way ANOVA determined significant differences between groups. Differences between means were tested by Tukey's least significant difference. Dates are presented as means and standard error of the means (SEM) and the animal was the experimental unit. It was performed a simple regression analysis to relate the differences caused in the fatty acid composition with the fatty acids consumption and total production of volatile fatty acids in *caecum* and proximal colon.

III –Results and discussion

In Table 2 the FA composition of subcutaneous fat and difference between initial and final period of experiment is shown. FA composition was not significantly different between diets when the experiment begins, but it was significantly different after 28 days the diets ingestion. In the total of saturated and polyunsaturated is where can be appreciate these differences.

Table 2. Fatty acid composition (g·100g^{-1} fat) of the subcutaneous adipose tissue and the difference between the fatty acid composition the initial and final experimental time

Diets	1	2	3	4	5	SEM	p-value
Subcutaneous							
∑Saturated	38.3 ^{b}	36.7 ^{ab}	34.6 ^{a}	36.0 ^{ab}	35.8 ^{a}	0.353	0.004
∑Monounsaturated	52.3	52.9	53.5	53.0	52.7	0.225	0.549
∑Polyunsaturated	8.7 ^{a}	9.4 ^{ab}	11.0 ^{c}	10.3 ^{bc}	10.6 ^{bc}	0.239	0.002
∑Odd-chain fatty acids	0.8	0.9	0.9	0.7	0.9	0.031	0.519
Difference							
∑Saturated	0.18 ^{b}	0.67 ^{b}	-1.95 ^{a}	-1.00 ^{ab}	-2.35 ^{a}	0.384	0.031
∑Monounsaturated	2.04	1.90	2.44	2.67	3.14	0.192	0.254
∑Polyunsaturated	-2.17 ^{ab}	-2.56 ^{a}	-0.51 ^{b}	-1.62 ^{ab}	-0.87 ^{ab}	0.242	0.017
∑Odd-chain fatty acids	-0.05	0.00	0.02	-0.04	0.07	0.019	0.239

Means with the different letters on the same line are significantly different.

Results of total volatile fatty acid and molar proportion in the caecum and proximal colon are shown in Table 3. Total VFA concentrations in the caecum and colon of pigs have been reported to be between 80 and 240 mmol·l^{-1} in several studies (Pierce *et al*., 2005; Reilly *et al*., 2010). Although the volatile fatty acids production is high (both total and propionic acid) there is no relation to odd-chain fatty acids composition (Table 4).

Table 3. Effect of experimental diets on total volatile fatty acid (mmol·l^{-1}) and on the molar proportion (%) in the caecum and proximal colon†

Diets	1	2	3	4	5	SEM	p-value
Caecum							
Total VFA (mmol•l^{-1})	299	291	249	228	295	10.41	0.090
Acetic acid	71.4ab	74.2b	69.7a	71.9ab	74.4b	0.54	0.011
Propionic acid	19.5ab	17.1a	22.1b	19.0ab	16.4a	0.57	0.003
Butyric acid	8.1	8.0	7.0	8.0	7.9	0.23	0.568
Proximal colon							
Total VFA (mmol•l^{-1})	317b	296ab	251ab	235a	308ab	9.93	0.011
Acetic acid	70.3	71.4	68.4	70.1	71.4	0.47	0.246
Propionic acid	18.3ab	18.2ab	21.7b	19.1ab	17.4a	0.50	0.045
Butyric acid	9.8	9.3	8.2	9.3	9.4	0.28	0.497

†Acetic, propionic, butyric, isobutyric, isovaleric and valeric acid. Means with the different letters on the same line are significantly different.

Fievez *et al*. (2003) suggested that cis-9 C17:1 was endogenously produced from C17:0 by Δ9-desaturase activity, consistent with a product/precursor relationship. In our case, the ratio of odd-chain fatty acids is narrow, with the adipose fat concentration of cis-9 C17:1 and C17:0 closely related (r^2=0.763).

The fat composition is determined by feed, either by direct diet lipids deposition or by de novo synthesis from carbohydrates ingested. For the monounsaturated and saturated, the fatty acids intake influences the composition. The correlation coefficients in the regressions analyses reflect the relation between consumption and differences in FA composition (Table 4). In the case of monounsaturated FA, this relationship is positive and negative in saturated.

Table 4. Regression analysis with correlations coefficients

	Difference			
	Saturated	**Monounsaturated**	**Polyunsaturated**	**Odd-chain**
Consumption†	-0.65*	0.49*	-0.10	0.42
Total VFA *caecum* (mmol/l)	0.37	-0.21	-0.42	-0.07
Total VFA colon (mmol/l)	0.22	-0.12	-0.27	0.24
Propionic *caecum* (mmol/l)	0.49*	-0.47*	-0.39	-0.21
Propionic colon (mmol/l)	0.34	-0.28	-0.33	0.14

†Total consumption of each fatty acid per day, respectively.
* Marked correlations are significant at $p < 0.05$

IV – Conclusions

The subcutaneous fatty acid composition varies depending on the type of diet given to pigs. Volatile fatty acids production not changes the composition of the odd-chain fatty acids.

Acknowledgments

The research was supported by the Regional Government Junta de Extremadura and the European Social Fund. Research (Project PRI08B091). The authors gratefully acknowledge to workers the farm Valdesequera for their technical assistance in animal management and Fanny Senie for her help in the laboratory.

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The fiber in the diet of Iberian pigs and ability to limit growth

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Abstract. In order to limit the growth of Iberian pigs during the rearing period, is to consider to include in the diet fibrous material, to reduce food intake under *ad libitum* feeding and thus energy. Incorporation levels are 300, 450 and 600 g kg^{-1}, using as fiber source, sugar beet pulp (as soluble fiber) and cereal straw (as insoluble fiber). A total of 20 pure Iberian pigs was used (into 5 groups of 4 pigs each). The pigs started the experiment with 84 kg of live weight and remains in the experiment for 4 weeks, to reach a live weight of 102 kg. Growth and feed intake was controlled and average daily gain and feed conversion was determined. The *ad libitum* intake of 300 to 600 g kg^{-1} of fiber produces a decrease in all the productive parameters except in feed conversion. The effect of the type of fiber over the feed intake was significant with B-coefficients of -0.039 for SBP and -0.052 for CS. The cereal straw has a major effect in the decrease of the intake than the sugar beet pulp.

Keywords. Fiber – Feed – Growth – Iberian pig.

Les fibres dans l'alimentation des porcs Ibériques et leur capacité à limiter la croissance

Résumé. *Afin de limiter la croissance des porcs Ibériques au cours de la période d'élevage, il est à envisager d'inclure des matériaux fibreux dans le régime alimentaire* ad libitum*, afin de réduire la consommation alimentaire, et donc l'énergie. Les niveaux d'incorporation sont de 300, 450 et 600 g kg*$^{-1}$*, en utilisant comme source de fibres, de la pulpe de betterave sucrière (fibres solubles) et de la paille de céréales (fibres insolubles). On a employé un total de 20 porcs Ibériques purs divisés en 5 groupes de 4 porcs chacun. L'expérience a commencé à 84 kg de poids vif, et les porcs sont restés pendant 4 semaines jusqu'à ce qu'ils atteignent un poids de 102 kg. L'ingestion de 300 à 600 g kg*$^{-1}$ *de fibres produit une diminution des paramètres productifs excepté l'indice de conversion alimentaire. Concernant l'effet du type de fibre, il a été observé que l'ingestion a été significative avec les coefficients de B (- 0,039) pour SBP et (-0,052) pour CS. La paille de céréale a davantage d'effet dans la diminution de la consommation que la pulpe de betterave.*

Mots-clés. *Fibres – Croissance – Porc ibérique.*

I – Introduction

The legislation of the products of the Iberian pig (RD 1469/2007 of November 2, 2007), that the animals must reach, for his entry in fatten or "montanera", a not excessively high weight (between 92 and 115 kg) and to comply a few minimal requirements of age of sacrifice and permanency according to type of finish fattening. To obtain this aim, from 23 kg and 3 months of life, up to ten-twelve months of age, there have to be obtained average daily gain of 270-440 g/día, really low for his potential of growth. It is necessary to offer a balanced and efficient diet (Barea *et al*., 2007) that produces animals with a good conformation at the conclusion of the phase of growth and with a suitable skeletal structure and muscular development to confront the finish fattening period.

The strategy used in the sector of the Iberian pig is to restrict the diet, administering of form rationed during the period of growth. The disadvantage of this system is in the managing, in which it is necessary to administer the feed in a ration daily, with the consequent workforce that it carries. Another disadvantage derived from the rationing is the low level of satiety reached, causing problems of behavior and inequality between them. To administer the diet *ad libitum* would be a solution but the Iberian pig have a high capacity of ingestion and a minor mean retention time of the digesta (Nieto *et al*., 2001; Morales *et al*., 2002), which is unviable for excessive average weight in period to entry in "montanera". It would be so interesting, to have a possibility to be able administer a diet *ad libitum*, with a low energetic concentration based on the use of fibrous raw materials in a high percentage and that produces a high satiety to give solution to this problem.

The aim is to study the possibility of including in the diet of growing Iberian pigs (84-102 kg of body weight) dietary fiber, for limit the energy consumption and administer the diet a*d libitum*.

II – Materials and methods

A total of 20 Iberian pigs barrows males divided in five lots was used. Sugar beet pulp SBP(as soluble fibre) and cereal straw CS (as insoluble fibre) commercially available were incorporated as the major fiber sources in diets according to the scheme presented in the Table 1. The diets were milled to pass through a 2 mm screen diameter and presented as granule. The chemical composition of the diets is presented in Table 1. The diets contained similar crude protein contents and total amino acids, calcium and phosphorus levels and were balanced for metabolizable energy (ME).

Table 1. Content of fiber sources (g kg^{-1}) and chemical composition (g kg^{-1} dry matter) on the experimental diets

Diets	1	2	3	4	5
Sugar beet pulp	150	300	150	225	300
Cereal straw	150	150	300	225	300
Crude protein	142	150	148	157	153
NDF†	280	350	382	407	420
ADF†	151	170	230	199	234
ADL†	17	21	25	22	27
Hemicellulose††	129	180	152	208	187
Cellulose††	134	148	206	177	207
ME (mcal kg^{-1} fresh matter) †††	2.45	2.40	2.35	2.39	2.33

†NDF: neutral detergent fiber, ADF: Acid detergent fiber ADL: acid detergent lignin.
††Hemicellulose and cellulose are NDF-ADF and ADF-ADL, respectively, according to Van Soest *et al*. (1991).
†††Calculated for tables FEDNA 2003.

The body weight (BW) of pigs were determined at the beginning and the end of the experiment and feed consumption was determined daily by weighing the feed offered and refused. Daily BW gain and feed conversion ratio were calculated. The pigs started the experiment with an average live weight of 84 kg and remained there for 28 days, up to 102 kg of live weight.

The experimental design of the investigation was the response surface methodology applied to a full factorial central composite design with two independent variables (amount of SBP and amount of CS) at two levels (150 and 300 g kg^{-1} levels) obtaining the corresponding response. Four replicates at the centre points were also considered.

Regression analysis of the data obtained and estimation of the coefficients of the regression equation was realised. The fit of the regression model attained was checked by the Correlation coefficients. The Unscrambler software (Version 9.7, Camo Software AS OSLO, Norway 2007) was used for regression and the graphical analyses of the data obtained.

One-way ANOVA determined significant differences between groups. Differences between means were tested by Tukey's least significant. Dates are presented as means and standard error of the mean (SEM) and the animal was the experimental unit. These statistical analyses were performed using SPSS (version 15.0).

III – Results

In the Table 2 are presented the performance results. A significant effect of experimental diet was observed between a level **5** of incorporation of fiber (600 g kg^{-1}) and level **1** (300 g kg^{-1}) with ($p < 0.05$). Feed intake was affected by diet, was observed a decrease with an increasing of dietary fiber. Equally was observed in the live weigth and in the average daily gain decreases with increase of fiber. The voluntary intake is depressed when fiber is increased in the diet for effect of satiety (Cole *et al*., 1971). The administration of fiber reduced the feed intake of the animals, and increased the weight of gastro-intestinal tract content (Anguita *et al*., 2006).

Table 2. Effect of dietary diet on performance of pigs in 28 days of experiment period

Diets	1	2	3	4	5	SEM	p-value
Live weight increase (kg)	23.0 ^{a}	19.4 ^{ab}	17.9 ^{ab}	17.1 ^{ab}	15.0 ^{b}	1.035	*
Feed intake (kg per day)	4.47 ^{a}	3.91 ^{ab}	3.71 ^{ab}	3.79 ^{ab}	3.10 ^{b}	0.149	*
Average daily gain (kg per day)	0.821 ^{a}	0.692 ^{ab}	0.638 ^{ab}	0.611 ^{ab}	0.536 ^{b}	0.037	*
Feed conversion ratio	5.7	5.7	6.4	6.3	5.9	0.301	ns

SEM. Standard error of the means.
Means with the different letters on the same line are significantly different * $p < 0.05$ ns: no significant.

The effect of type of fiber is presented in Table 3. The feed intake was significant with B-coefficients of -0.039 for SBP and -0.052 for CS. This indicate that the straw has a major effect in the decrease of the intake that the sugar beet pulp. About the average daily gain, though the model is adjustment, was observed that the effect of SBP is not significant but the effect of straw is it ($p = 0.031$). All this is illustrated in Fig. 1 where are presents the effect of the factors amount of SBP and CS for feed intake (a) and average daily gain (b). It showed clearly that the inclination of the planes is different according to the source of fiber.

Table 3. Regression model fit with parameter estimates and correlations coefficients

		B-coefficients	p-value	Correlation coefficients
DFI	Model		0.006	0.675
	Intercept	3.796	0.000	
	SBP (A)	-0.039	0.037	
	CS (B)	-0.052	0.008	
ADG	Model		0.036	0.569
	Intercept	0.659	0.001	
	SBP (A)	-0.008	0.126	
	CS (B)	-0.011	0.031	

This result is in disagreement with other studies that the inclusion of sugar beet pulp in the diet reduced the voluntary feed intake of pigs more than other less digestible ingredients (Brouns *et al*. 1991). The type of fiber cause different effect in digestive process, saciety and digesta transit time (Caspar *et al*., 2001). The effect of saciety is associated to high amount of soluble dietary fibre which causes an increased volume of digesta. Is probably that the determinant factor limiting ingestion is the water retention capacity of soluble fiber (Kyriazakis *et al*., 1995). The effect of genotype of Iberian pig was contrasted for feed intake and digestibility coefficients (Morales *et al*., 2002) but there are not sufficient studies of digestive process of fiber and transit time in Iberian pig.

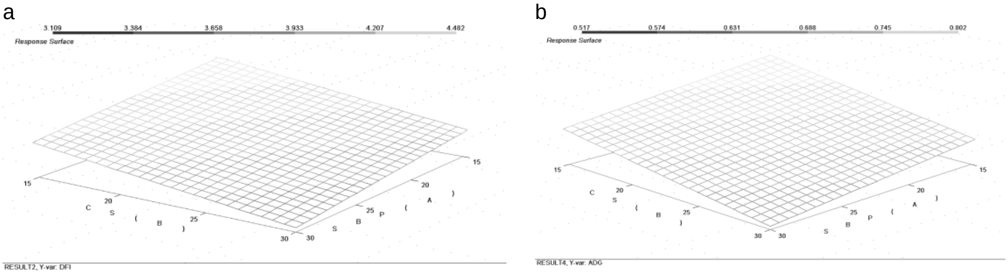

Fig. 1. Response surface plot of the effect of sugar beet pulp and cereal straw on the daily feed intake (a) and average daily gain (b).

IV – Conclusions

The results of the present study show that an increase of fiber in the diet produces a decrease in the productive parameter except in feed conversion. Is more efficient to control the capacity of ingestion of the Iberian pig and decrease his potential of growth at least 60 % of level of incorporation of fiber with a similar feed conversion that other percentage of incorporation. In parallel, the straw has a major effect in the decrease of the intake that the sugar beet pulp.

Acknowledgments

The research was supported by the Regional Government Junta de Extremadura and the European Social Fund. Research (Project PRI08B091). The authors gratefully acknowledge to workers the farm Valdesequera for their technical assistance in animal management.

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Assessment of the "recebo with postre" system

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Abstract. The aim of this work is to assess the quality of carcass and meat of pigs fed in free range with feed supplemented (system called "recebo" with "postre"). 205 pure Iberian pigs (18 months old with and an average weight of 111.6 kg at the beginning of the experiment) were used. A group of 102 pigs remained in free range for 85 days on a farm of 126.8 ha (Montanera: MN). Another group of 103 pigs were fed in other farm of 122.5 ha in free range and supplemented with concentrates (with 6.44% of crude fat and 59.3% of oleic acid) at a rate of 983 g per animal/day, for 69 days (Recebo: RC). Once slaughtered, measures were taken in the carcass and composition studies were performed on subcutaneous and intramuscular tissue (20 pigs per group). RC animals have better yields of primal cuts (32.2% for total of hams, shoulders and loins) than MN (28.9%). The *longissimus dorsi* intramuscular fat is the same statistically in RC (8.41%) than in MN (8.55%). The meat tocopherol composition reflects the diet: α-tocopherol: 4.78 and 2.02 mg/kg and γ-tocopherol: 0.47 and 1.07 mg/kg for RC and MN, respectively. The subcutaneous fatty acids are similar except oleic acid (54.07% in RC and 56.16% in MN).

Keywords. Iberian pig – Quality – Free range – Feed.

Evaluation du système de "recebo" avec "postre"

Résumé. *L'objectif du travail est d'évaluer la qualité de la carcasse et de la viande des porcs nourris en plein air avec des aliments supplémentés (système appelé "recebo" avec "postre"). 205 porcs purs Ibériques (18 mois et un poids moyen de 111,6 kg au début de l'expérience) ont été utilisés. Un groupe de 102 porcs est resté en plein air pendant 85 jours sur une ferme de 126,8 ha (Montanera: MN). Un autre lot de 103 porcs a été nourri dans une ferme de 122,5 ha en plein air et complémenté avec des concentrés (avec 6,44% de matières grasses brutes et 59,3% d'acide oléique) à un taux de 983 g par animal / jour, pendant 69 jours (Recebo: RC). Une fois abattus, des mesures ont été prises dans la carcasse et des études de composition sur les tissus sous-cutanés et intramusculaires ont été réalisées (20 porcs par groupe). Les animaux RC ont présenté de meilleurs rendements pour les parties nobles (32,2% pour l'ensemble des jambons, épaules et côtes) par rapport aux MN (28,9%). La graisse intramusculaire du* Longissimus dorsi*, est la même sur le plan statistique chez RC (8,41%) et MN (8,55%). La teneur de la viande en tocophérol reflète l'alimentation: α-tocophérol: 4,78 et 2,02 mg / kg et γ-tocophérol: 0,47 et 1,07 mg / kg pour les RC et MN, respectivement. Les acides gras sous-cutanés sont semblables, sauf l'acide oléique (54,07% chez RC et 56,16% chez MN).*

***Mots-clés.** Porc Ibérique – Qualité – Libre parcours – Alimentation animale.*

I – Introduction

In the traditional way of raising Iberian pigs under free range conditions (called "montanera"), the fattening period is completed with only the natural resources offered by the dehesa (mainly grass and acorns). Sometimes, it supplements the diet of these animals is supplemented with compound feed, and the system becomes "recebo". In the called "recebo" with "postre" the compound feed is added while animals remain in free range. In the other type, it is added when dehesa resources are depleted and continues until slaughter weight is reached. With "recebo" can be increased the number of pigs per hectare and primal cuts percentages. But this practice can result in a decreased quality of Iberian product. This paper compares the carcass and meat quality of montanera against "recebo" with "postre".

II – Materials and methods

A total of 205 Iberian pig (male and female castrated) and 18 months old was used. The pigs were divided in two groups: Montanera (MN) with 102 animals remaining in a farm of 126.8 hectares, fed on grass and acorns mostly and Recebo (RC) with 103 animals that were raised under free-range conditions with partial replacement of acorns and grass of concentrate (983 g/animal/day; 6.44% crude fat content and 59% of oleic acid) in another farm of 122.5 hectares.

After slaughtering, the hot carcass weights were recorded. The backfat thickness was measured in the middle line at the last and tenth rib level. The carcasses were cut and hams, shoulders and loins were weighed after trimming. The rest of the carcasses were separated into lean, bone, head, ribs, spine and fat. In 20 animals of each group was taken a subcutaneous tissue sample and a piece of *longissimus dorsi* muscle.

The intramuscular fat was calculated by the method of Bligh and Dyer (1959) (with minor modifications). The fatty acid composition was determined by gas chromatography after acid transesterification in the presence of sulphuric acid (5% sulphuric acid in methanol) (Cava *et al.*, 1997). The results are expressed as percentage of total fatty acids and analyzed a total of 11 fatty acids. α-tocopherol and γ-tocopherol in muscle were determined following the method described by Rey *et al.*, 1996.

The chemical, fatty acids and vitamin E composition of the grass and acorns are presented in Table 1. Throughout the fattening period, oak acorns were collected randomly and samples of grass were cut. The lipids were extracted with chloroform/methanol (1:2) according to the method described by Bligh and Dyer (1959) (with minor modifications). The food was analysed in fatty acid composition and vitamin E content with same methods than samples from animals. The chemical composition of acorns and grass was performed according to standard methods (AOAC, 1990).

Table 1. Chemical (g/100g dry matter), fatty acids (g/100g fatty acids) composition and vitamin E (mg/kg) content of the grass and acorns

	Acorn		Grass	
	Montanera	**Recebo**	**Montanera**	**Recebo**
Chemical composition, (g/100g dry matter)				
Crude fat†	8.41	7.87	4.62 (34.5)	4.34 (37.9)
Crude fiber	1.40	2.23	25.42	22.30
Crude protein	5.13	4.24	17.82	19.48
Fatty acids, (g/100g fatty acids)				
Palmitic (C16:0)	15.01	15.02	26.8	26.5
Estearic (C18:0)	3.99	4.38	5.3	6.1
Oleic (C18:1 n-9)	61.10	60.69	8.1	13.1
Linoleic (C18:2 n-6)	16.12	15.87	14.3	11.3
Linolenic (C18:3 n-3)	1.02	0.98	38.4	36.3
Vitamin E, (mg/kg)				
α-tocopherol	6.37	8.24	533.1	330.6
γ- tocopherol	41.85	39.82	-	-

†In parentheses % true fat.

All data were analyzed by one-way analysis of variance using the General Linear Model of SPSS, 2008 statistical software (v.15.). An individual pig was the experimental unit for analysis of all data. Data were expressed as the mean and deviation standard of each group together with the significance levels of the effect.

III – Results and discussion

Table 2 contains the information about growth performance obtained in pigs and all data yields and carcass composition of slaughter.

Despite having lower weight at slaughter, the hams, shoulders and loin of RC pigs have higher weights than those of MN. This is because the percentage of primal cuts increases as decreases carcass fat. Feeding in free range fattening phase is very unbalanced, with a large deficit of protein (Nieto *et al*., 2002). MN carcass are fatter than RC carcass.

Table 2. Growth performance, yields and carcass composition

	Recebo		Montanera		Significance
	Means	sd	Means	sd	
Initial weight, kg	112.6	12.56	110.7	10.26	ns
Final weight, kg	162.4	15.89	172.0	16.75	*
Weight gain, kg	49.8	8.75	61.3	11.01	*
Fattening days	69		85		
Average daily gain, g	721.8	126.81	722.1	149.73	ns
Hot carcass weight, kg	130.0	12.72	139.0	13.52	*
Fat depht, cm					
Last rib	5.6	0.76	6.9	0.93	*
Tenth rib	7.2	0.92	8.4	0.88	*
Primal cuts weight, kg					
Ham	22.8	2.36	22.1	1.89	ns
Shoulder	15,3	1,54	14,4	1,37	*
Loin	3.77	0.28	3.45	0.15	*
Carcass components, %					
Ham	17.5	0.85	16.0	0.85	*
Shoulder	11.8	0.53	10.4	0.66	*
Loin	2.9	0.31	2.5	0.26	ns
Lean	6.6		6.1		-
Bone, head, spine y ribs	5.9		5.7		-
Fat	55.3		59.3		-
Carcass composition†					
Lean, %	21.08		18.97		-
Fat, %	69.92		72.56		-

†According to data from percentage of fat and lean of ham and shoulder, Mayoral *et al.* (1999). *$p < 0.05$; ns: no significant.

The fatty acid compositions of the subcutaneous adipose tissue are shown in Table 3. The fatty acid composition of subcutaneous fat is one the available parameters to classify Iberian pigs based on their food, which is especially important. RC pigs differ from MN in monounsaturated acids.

Table 3. Fatty acid composition of subcutaneous adipose tissue (g/100 g total fatty acids)

	Recebo		Montanera		
	Means	sd	Means	sd	Significance
Palmitic (C16:0)	19.05	0.72	18.93	0.89	ns
Estearic (C18:0)	9.25	0.67	874	0.96	ns
Oleic (C18:1 n-9)	54.07	1.04	56.16	1.32	*
Linoleic (C18:2 n-6)	9.96	0.56	9.26	0.51	ns
Linolenic (C18:3 n-3)	1.32	0.26	0.88	0.10	*
∑ Saturated	30.3	1.21	29.5	1.71	ns
∑ Monounsaturated	58.4	1.11	60.4	1.39	*
∑ Polyunsaturated	11.3	0.76	10.1	0.58	*

* $p < 0.05$, ns: no significant.

Since the animals begin the free range, the percentage monounsaturated fatty acid increases in detriment of polyunsaturated fatty acid, mainly due to the fat composition of the acorn. But only significant differences in oleic and linoleic fatty acids are found. The percentage of oleic acid is significantly higher in MN pigs, due to greater availability of acorns and longer fattening period. Linolenic acid is higher in RC group, surely the reduced availability of acorns caused the animals to consume more grass, the main provider of this fatty acid. We can say that there is no differences comparing the fatty acids percentages in RC pigs with those obtained by different authors in Iberian pig classified as montanera (Daza *et al*., 2005).

Table 4 shows two representative parameters of meat quality, intramuscular fat and vitamin E in muscle. Despite having more carcass fat and more time in the fatting period, MN pigs do not show a higher percentage of intramuscular fat than RC.

Table 4. Intramuscular fat and vitamin E content of longissimus dorsi muscle

	Recebo		Montanera		
	Means	sd	Means	sd	Significance
IMF, %†	8.46	2.28	8.55	3.31	ns
α-tocopherol, mg/kg	4.78	0.92	2.02	0.60	*
γ- tocopherol, mg/kg	0.47	0.08	1.07	0.27	*

†Intramuscular fat.
*$p < 0.05$, ns: no significant.

With regard to vitamin E, we can say that every type of feeding give a different amount in α-tocopherol and γ- tocopherol. RC pigs have higher α-tocopherol levels than MN pigs, while the latter have higher levels of γ- tocopherol. Although the grass eaten by RC pigs had a lower amount of α-tocopherol than that of MN pigs and the feed supplemented contained small amounts of α-tocopherol, RC pigs have higher levels of α-tocopherol. This is because they consume more grass, probably because of the scarcity of acorns they had to satisfy their appetite with grass. MN pigs have more than double the amount of γ- tocopherol than RC pigs because they consumed more acorns (acorns is the only food that could provide γ- tocopherol to pigs) (Rey *et al*., 2006).

IV – Conclusions

When supplemented with concentrate (with specific amounts and certain qualities), Iberian pigs reared under free-range conditions (called "recebo") can improve carcass composition without reducing the meat quality. The γ-tocopherol content in meat could differentiate between acorns levels of consumption.

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Soil degradation by grazing pig in Mediterranean environment

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Abstract. The breeding of pigs grazing can cause serious environmental problems for the particular feeding behaviour of these animals. The aim of this research was to study the soil damage caused by pigs grazing in a Mediterranean forest. Eight adult pigs were introduced in an enclosure of 1300 m2 (with a density of about 200 m2 per animal), partially wooded. Experimental observations were made about visual characterization of damage, physical and chemical characteristics of soil structure. The results showed an initial compaction of the enclosure perimeter with a progressive increase of the rooting action and excavation over the entire surface. Particularly, the excavation damages phytocoenosis because causes the destruction of turf and tree and shrub roots. Soil organic matter increases after a short grazing period (increase of C organic and N total), while it decrease at the end of the trial. Soil physical characteristics showed a worsening during grazing period, with a progressive loss of porosity and soil structure. All of these results showed that soil damages caused by pigs grazing are extremely serious with negative repercussion on the entire ecosystem.

Keywords. Pig – Soil – Environment – Organic matter.

La dégradation des sols par le pâturage de porcs dans l'environnement méditerranéen

Résumé. *L'élevage de porcs au pâturage peut entraîner de graves problèmes d'environnement dû au comportement alimentaire particulier de ces animaux. L'objectif de cette recherche était d'étudier la dégradation des sols causée par les porcs au pâturage dans une forêt méditerranéenne. Huit porcs adultes ont été introduits dans un enclos de 1300 m^2 (avec une densité d'environ 200 m^2 par animal), en partie boisé. Les observations expérimentales ont été faites sur la caractérisation visuelle des dégâts et sur les caractéristiques physiques et chimiques de la structure du sol. Les résultats ont montré un compactage initial du périmètre avec une augmentation progressive de l'action de fouissage et d'excavation sur toute la surface. En particulier, l'excavation endommage la phytocénose par la destruction du gazon et des racines des arbres et des arbustes. La matière organique du sol a augmenté après une courte période de pâturage (augmentation de C organique et N total), tandis qu'elle diminue à la fin du processus. Les caractéristiques physiques du sol ont montré une détérioration durant la période de pâturage, avec une perte progressive de la porosité et de la structure du sol. Tous ces résultats ont montré que les dommages du sol causés par des porcs au pâturage sont extrêmement graves, en raison des incidences négatives sur l'écosystème tout entier.*

Mots-clés. *Porc – Sol – Environnement – Matière organique.*

I – Introduction

Due to some external agents of interference, the soil may undergo a degradation process which causes a regression from a high quality level to a low quality one; furthermore, in case of an extreme desertification it may cause the total loss of the soil's biological potential and its resilience. Among the possible causes of this desertification process, the most important one is the high animal pressure. In fact, the animals impact on the soil degradation both in a direct way and in an indirect one. The pigs grazing may cause a reduction of the vegetation which protects the soil from an erosion process and it may guarantee a good level of organic matter. A further effect of pigs grazing is the soil compaction caused by pigs trampling. Both those reasons may lead to a reduction of the soil's vitality and fertility also showed through chemical and physical structural parameters. Among livestock animals, pigs cause the most relevant environmental problems for their specific behavioural characteristics.

The two main damaging effects on the soil are the rooting and the soil compaction. The rooting is based on pigs excavation activity to find tubers, roots, and terriculous phauna. Furthermore, it may reduce the surface layers of soil of about 20-30 cm causing a high reduction of the vegetable biomass (so, the involved areas look like a milled soil), (Barrett, 1982; Singer *et al.,* 1984). The soil compaction is due to pigs habit to pass through selected paths; in this way, it stops the vegetation growing and the soil becomes asphyxial.

The aim of this research was to check the time evolution of the soil's quality and turf exposed to pigs overgrazing in a specific forest area.

II – Materials and methods

The research took place in a farm, (Pisa, Italy) located in woodland typical of the Mediterranean coast. For the trial was fenced a flat pasture area of about 1300 m^2 with a high density of animals (160 m^2 per animal). The area was representative of the forest, and was never used before as grazing land. Eight "Large White" pigs were entered in this area, according to a continuous grazing technique, and fed with organic diet. Environmental monitoring was carried out for a period of 100 days long. Four sampling were carried out after 0 (T0), 15 (T1), 45 (T2) and 100 (T3) days, to evaluate quantitative and analytical soil's damage.

The observations (expressed as percentage of disturbed area) were made to determine soil's direct damage (trampling and rooting activity); moreover, the over 5 cm depth wallows were measured, to evaluate the surfaces and volumes of removed soil. On the samples analysis focused on chemical and physical-structural parameters were carried out. Three samples per plots of $1m^2$, were collected for each type of damage and each time of sampling, according the follow scheme: a) control (undisturbed soil) at T0; b) two types of soil damage: rooting activity and trampling activity at T1, T2, T3.

For the trial analyzed the following analytical parameters have been:

Bulk Density assessed according Blake and Hartge (1986) method; porosity evaluated using the bulk density according the following formula: P=1-(BD/PD)/100, where BD= Bulk Density and PD= Real Density (=2.65 g/cm^3 for a medium texture soil). Cracking, analyzed according Petruzzelli and Guidi (1975) method. Total Organic Carbon (TOC), evaluated by an elemental analyzer RC-412 MULTIPHASE CARBON, and Total Nitrogen (TN) assessed by an elemental analyzer FP-528 PROTEIN/NITROGEN DETERMINATOR.

All results are the means of determination made on three replicates. To evaluate the effect of damage, the chemical and physical parameters were analyzed by ANOVA one way, within each treatment (rooting and trampling activity). The significant level reported ($P < 0.05$) are based on the Pearson coefficients.

III – Results and discussion

The qualitative assessment of environmental damage, directly observed on the soil can be summarized in Table 1.

At T1, the presence of a trampled area along the fence and a rooting activity which interested one third of the surface was detected. Moreover, a sporadic wallowing activity was revealed.

At T2, trampling activity spread not only in the peripheral zone but also in the inner pasture area. Much of the surface was rooting and contemporary the excavated area percentage increased.

At T3 is not possible to distinguish the two different types of effects because the surface is totally degraded by rooting activity, so the last sampling was done only on the soil rooting. The wallows were widespread.

Table 1. Soil damage

	T0 (0 days)	T1 (15 days)	T2 (45 days)	T3 (100 days)
Trampling activity (%)	0	10	25	0
Rooting activity (%)	0	30	65	100
Undisturbed (%)	0	60	15	0
Wallowing activity (m^2)†	0	5.8	12.85	18.12
Wallowing activity (m^3)††	0	0.5	1.32	1.69
Wallows (n)	0	5	12	18

†Area estimated as ellipse.
††Volume estimated as semi ellipsoid: (4/3*a*b*c*3.14)/2

The organic matter content, expressed as Total Organic Carbon (TOC%) and Total Nitrogen (TN,%) (Fig.1 A, B), decreases during the trial in soils subjected to rooting activity, while increases in trampled soils. These parameters change significantly over time, this is a negative indicator for the estimation of soil fertility.

In the case of high animal density, rooting causes the complete removal of biomass in the soil (roots, leaves, small branches, etc.), which is partly eaten by animals and does not restore the soil organic matter sink. Furthermore, excessive compaction reduces roots and plants growth, preventing the restoration of soil organic matter content.

Along the trial, in trampled soils, the increase of both parameters could be due to continuous grazing that led to an accumulation of manure and alter the true values of TOC and TN in the soil. Moreover, the decomposition of crop residues typical of the forest, may have contributed to increase both parameters.

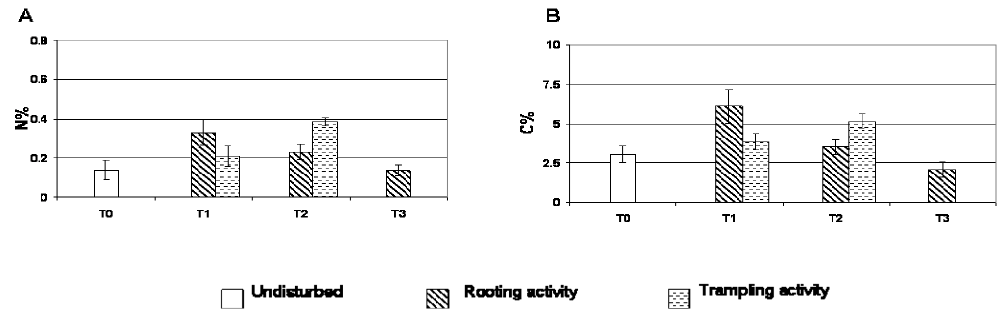

Fig. 1. A) Total Nitrogen (TN, %) B) Total Organic Carbon (TOC, %).

The bulk density parameter (Fig. 2, B) is inversely related to soil porosity (Fig. 2, A), which expresses the volume of voids of the soil as the percentage of the total volume. This physical property directly affects the dynamics of the gaseous and liquid phase in soil and, indirectly, the chemical fertility. Moreover, it shows a close correlation with the structure and tillage. The activity of overgrazing by pigs caused a worsening of soil porosity (increase in bulk density), but the intensity of this disturbance is greater in the trampled soil, due to excessive trampling by animals ($P > 0.05$). This effect in rooting soil is less evident.

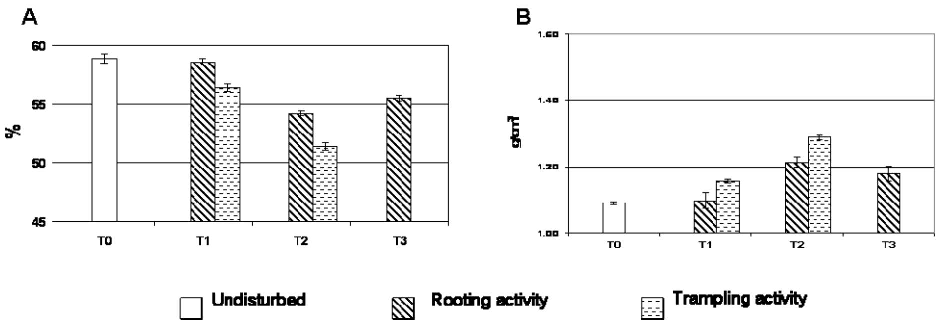

Fig. 2. A) Porosity (%) B) Bulk Density.

In rooting soil there is an increase of total cracking (Fig. 3, C) in relation to undisturbed soil, a value that decrease from T1 to T3 ($P<0.05$).

In particular this increase at T1 is characterized by a small number of cracks with a diameter greater than 500 µm (Fig. 3, B) [a dimension class of cracks representing the micro-habitat for soil microorganisms and, thus, the seat of microbiological and biochemical processes (Guidi *et al*., 1978)], but a high content of cracks with a diameter less than 500 µm (Fig. 3, A) which correspond to micro-porosity, can be regarded as a reservoir to hold water for plants and microorganisms (Pagliai *et al.,* 1980).

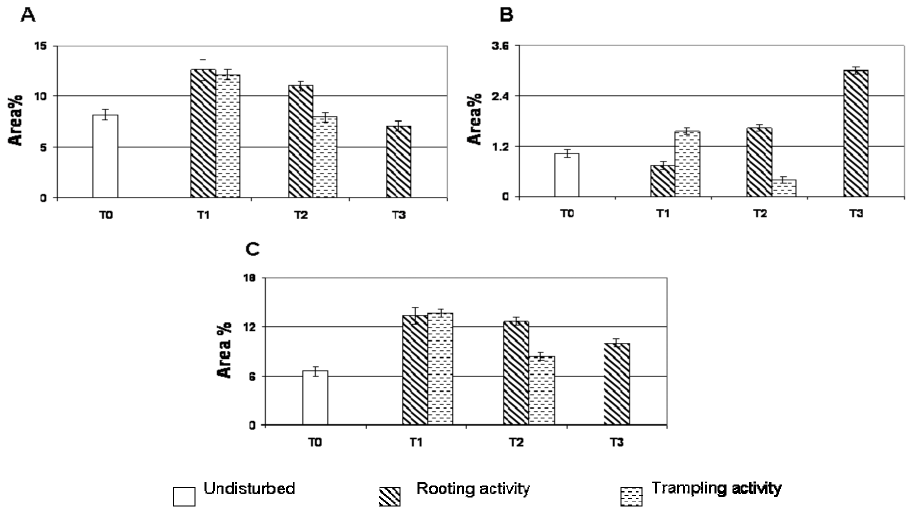

Fig. 3. A) Cracks <500 µm. B) Cracks >500 µm. C) Total cracks.

A mild rooting activity resulted in a beneficial effect on the microporosity of the soil but it causes adverse effects on the class cracks that are the habitat of soil microorganisms leading to a constraint normal course of microbial activity (Masciandaro *et al.,* 1997).

At the second sampling according with an evident rooting activity, the trend is changed. The cracks >500 μm are more than those <500 μm. This is a positive phenomena as far as a chemical field is concerned, but negative in relation to the soil structure. In trampled soil at T1 both the total cracks and the two ranges of cracks increase, according with the decrease of porosity, while at T2 both the total cracks and the two ranges of cracks decrease. This indicates that poaching has caused the soil compaction.

IV – Conclusions

The problem of damage to the soil by pigs is a significant environmental issue. Macroscopic aspects and also less visible factors directly affect the quality of soil grazed by pigs and indirectly the functionality of the entire ecosystem. Even in a limited period the animals degrade the soil's quality. The results, in fact, show as the soil's damage (soil removal caused by rooting and trampling) is closely related to the analytical small scale parameters on the physical and chemical characteristics of soil (cracks, bulk density, organic matter) .

After just over a month the soil is severely compromised in its main structural features. The high compaction by animals creates an asphyxial condition and compromises the soil fertility. The results show that a slight grazing by pigs and the moderate stirring of the surface layers of soil don't give problems on the soil. Therefore, we must be careful in pig density and animal farming system used that must be rational and proportionate to the type of environment and food availability in grazing area. Should be absolutely avoided continuous grazing, adopting a grazing round to make shifts on the pasture and to protect the soil characteristics.

Acknowledgements

The authors wish to thank Dr. G. Conte for technical assistance. Dr. S. Doni, Dr. E. Peruzzi and Dr. C. Macci for their intellectual contribution.

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Reproduction performance parameters in Iberian pig farms

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Abstract. Technical efficiency of the production is very important for cost control. As an industry, swine producers need to collect production data to use these to make decisions on their farms. Producers need benchmarks to compare their farm performances. There are few reproduction performance parameters about Iberian pigs. A total of 8 farms and 3400 sows providing data in a routine have been included in this first study summarizing 2009 results. Data were imported from different software brands, uniformized and merged in a single data base, from which the analysis was performed using Pigchamp© software, obtaining the following mean values: weaning to service interval = 8,4 days, farrowing rate = 75 %, returns to estrus = 18,4 %, total born = 8,3, born alive = 7,8, weaned piglets = 7, number of litters per sow and year = 2,11, piglets weaned per sow and year = 14,4. The aim of this paper is to provide a first structured batch of the most important reproductive parameters.

Keywords. Iberian pig – Productivity – Benchmarking.

Paramètres de rendement reproductif dans des fermes de porc Ibérique

Résumé. *L'efficacité technique de la production est très importante pour le contrôle des coûts. Comme dans toute industrie, les producteurs de porcins ont besoin de recueillir des données productives pour la prise de décisions dans leurs fermes ; et ils ont besoin de paramètres de référence ("benchmarks") avec lesquels comparer le rendement de leurs fermes. Aujourd'hui, il y a peu de données sur l'efficacité reproductive du porc Ibérique. Dans ce travail on analyse les résultats de 2009 pour un total de 8 fermes et 3.400 truies incluses dans une routine de recueil et d'analyse de données. Ces données ont été importées de différents logiciels de gestion, elles ont été standardisées et consolidées dans une base de données qui a été analysée avec le logiciel Pigchamp© de gestion de fermes porcines, pour obtenir les valeurs moyennes suivantes : intervalle sevrage-saillie = 8,4 jours, taux de mise-bas = 75%, retour en œstrus = 18,4%, nés totaux = 8,3, nés vivants = 7,8, cochons sevrés = 7, nombre de mises-bas par truie et année = 2,11, cochons sevrés par truie et année = 14,4. Le but de ce travail était de fournir un premier rapport sur les principaux paramètres reproductifs.*

Mots clés. *Porc Ibérique – Productivité – "Benchmarking".*

I – Introduction

Technical efficiency of the production is very important for cost control. As an industry, swine producers need to collect production data to use these to make decisions on their farms. Producers need benchmarks to compare their farm performances. Iberian pig is a non improved rustic breed and there is a lack of reproduction performance parameters about it. The aim of this paper is to provide a first structured batch of the most important reproductive parameters as tool for benchmarking.

II – Materials and methods

1. Animals

A total of 8 farms and 3400 Iberian breed sows were studied.

2. Measures and analysis

This first study summarizes the production results of 2009. Data were imported from different software data bases, uniformed and merged in a single data base in order to analyze it with the Pigchamp© software. Overall approach was based in the differential diagnosis proposed by Dial *et al.* (1992) and the split of non productive days was based in the six components proposed by Koketsu (2005).

III – Results and discussion

Figure 1 shows the tree of productivity, with the following mean values: weaning to service interval (WSI) = 8.4 days (July and August were the months with the highest WSI and December was the one with the lowest WSI, these were 10 and 6.7 days respectively); farrowing rate (FR) = 75% (according to mating month, May had the highest FR and August had the lowest FR, these were 86.8 and 61.1% respectively); returns to estrus = 18.4%; total born = 8.3; born alive = 7.8; weaned piglets = 7; number of litters per sow and year = 2.11; and piglets weaned per sow and year = 14.4.

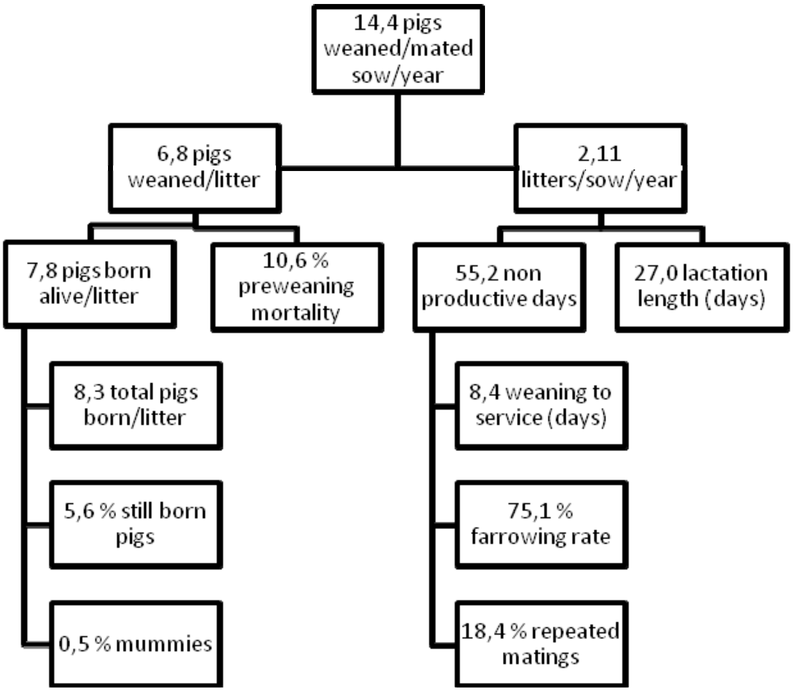

Fig. 1. Tree of productivity for the Iberian breed.

According to the pig industry standards (i.e. Carr, 2004) the FR is low (75% *vs* <82 to consider it a problem to improve), the WSI is high (8.4 days *vs* >7 to consider it a problem to improve), and the number of litters per sow and year is too low (2.11 *vs.* <2.3 to consider it a problem to improve). The number of litters per sow and year is clearly affected by the very high number of non productive days (Koketsu, 2005).

The WSI and FR monthly differences show a clear seasonal pattern, where the hot summer affects the reproductive performances.

The prolificacy performances are explained by the physiological differences of the Iberian breed.

IV – Conclusions

These results show an overall different reproductive performance with other breeds that can be explained both for the physiological differences and the management. The knowledge derived from this data should be used to compare farms and to establish objectives of reproduction performance.

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Influence of farrowing number on prolificacy and lactating performance in Iberian pig farms

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Abstract. Productivity of sows depends on farrowing number. Swine producers need to know how their sows produce to make decisions on culling and replacement besides cross-fostering strategies in their farms. Producers need benchmarks to compare their farm performances. There are few reproduction performance parameters about Iberian pigs. A total of 8 farms and 3400 sows providing data in a routine have been included in this first study summarizing 2009 results. Data were imported from different software brands, standardized and merged in a single data base, from which the analysis was performed using Pigchamp© software, obtaining performances according to farrowing number. Sows of 3rd and 5th farrows are the most prolific (8,2 born alive piglets) and sows of 3rd farrow wean more (7,2 weaned piglets); while sows of 1st, 2nd and ≥7th farrows produce less born alive piglets than average and 1st and ≥6th farrows wean less piglets than average. Hence 6th farrow should be considered to cull the majority of sows.

Keywords. Iberian pig – Productivity – Farrowing.

Influence du numéro de mise-bas sur la prolificité et le nombre de sevrés dans des fermes de porc Ibérique

Résumé. *La productivité de la truie dépend de son numéro de cycle ou mise-bas. Les producteurs de viande porcine ont besoin de savoir quelle est la production de leurs truies pour la prise de décisions concernant l'abattage et le renouvellement, et pour les stratégies de cession et d'adoption de cochons de lait dans leurs fermes. Les producteurs ont besoin de paramètres ("benchmarks") avec lesquels ils puissent comparer le rendement de leurs fermes. Aujourd'hui il n'y a pas beaucoup d'information sur les paramètres reproductifs du porc Ibérique. Dans ce travail on analyse les résultats de 2009 pour un total de 8 fermes et 3.400 truies incluses dans une routine de recueil et d'analyse de données. Ces données ont été importées de différents logiciels de gestion, elles ont été standardisées et consolidées dans une base de données qui a été analysée avec le logiciel Pigchamp© de gestion de fermes porcines, pour obtenir les rendements selon le numéro de mise-bas. Les truies en 3^e et 5^e mises-bas sont les plus prolifiques (8,2 cochons nés vivants) et celles en 3^e mise-bas sont celles qui ont sevré le plus d'animaux (7,2 cochons sevrés); tandis que les truies en 1^e, 2^e et ≥7^e mises-bas ont moins de cochons nés vivants que la moyenne, et celles des 1^e et ≥6^e mises-bas sèvrent moins de cochons que la moyenne. C'est pourquoi la 6^e mise-bas devrait être considérée comme le moment de l´abattage pour la plupart des truies.*

Mots-clés. *Porc Ibérique – Productivité – Portée.*

I – Introduction

Technical efficiency of the production is very important for cost control. As an industry, swine producers need to collect production data to use these to make decisions on their farms. Producers need benchmarks to compare their farm performances. Iberian pig is a non improved rustic breed and there is a lack of reproduction performance parameters about it. The aim of this paper is to provide a first structured batch of the productivity pattern according to the farrowing number as tool for benchmarking.

II – Materials and methods

1. Animals

A total of 8 farms and 3400 Iberian breed sows were studied.

2. Measures and analysis

This first study summarizes the production results of 2009. Data were imported from different software data bases, uniformed and merged in a single data base in order to analyze it with the Pigchamp© software.

III – Results and discussion

Reproductive performance of this pig breed is much lower than other modern pig lines; this would be explained by the physiological differences of the Iberian breed.

The total number of piglets born and born alive is very short (7.8 born alive and 6% of still born), besides preweaning mortality is not particularly low (9.7 %), resulting only 6.8 weaned piglets (Table 1).

Table 1. Distribution of farrowing number and productivity for the Iberian breed

Farrowing number	1	2	3	4	5	6	≥7	Average
Census (%)	15	20	16	14	15	7	12	-
Piglets born alive	7.1	7.5	8.2	8.1	8.2	8.1	7.7	7.8
Still born (%)	6	6	4	4	5	8	8	6
Weaned piglets	6.5	6.9	7.2	7.0	6.9	6.5	6.2	6.8
Dead piglets	1.3	1.2	1.3	1.2	1.4	1.6	1.3	1.0
Dead piglets (%)	17.3	12.7	11.0	19.3	10.5	14.0	7.6	10.6

The census distribution lacks of a higher per cent of primiparous as it is recommended (only 15 % of the census); hence replacement gilts were included in a lower percentage to the proper needs.

Prolificacy curves show a similar pattern compared to modern breeds. So sows of 3rd and 5th farrows are the most prolific (8.2 born alive piglets) and sows of 3rd farrow wean more (7.2 weaned piglets); while sows of 1st, 2nd and ≥7th farrows produce less born alive piglets than average and 1st and ≥6th farrows wean the same number of piglets but less than average.

IV – Conclusions

These results show an overall different reproductive performance with other breeds; however the Iberian breed follow a similar productivity pattern according to the farrowing number. As 6th farrowing sows wean as first farrowing sows, the 5th or 6th farrows should be considered to cull the majority of sows.

The knowledge derived from this data should be used to compare farms and to establish objectives of reproduction performance.

Session 3
Production systems – Nutrition

Effect of number of utilizations in a barley grass in Mediterranean conditions on biomass production and the admissible stocking rate of Majorcan Black Pig

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Abstract. Forages have significant economic value as a food resource for the Majorcan black pig and their environmental and social value is widely recognized. The multifunctionality of livestock production is due to its economic, ecological and social importance and is fully linked to its sustainability. The Majorcan black pig production has been directly linked to the exploitation of agricultural production. The better use of forages has a clear economic impact on the Majorcan black pig production and hence its sustainability. We have studied the biomass production and assessed the Majorcan black pig stocking rate of a barley grass, depending on the number of uses. We used an experimental plot with 4 treatments and three replicates per treatment. Under the agronomic conditions of our study, increasing to three the number of uses, allow more biomass, more protein, more energy, more servings and therefore more stocking rate of Majorcan black pig than two or one use.

Keywords. Majorcan black pig – Sustainability – Number of uses of forages.

Effet du nombre d'utilisations d'un pâturage d'orge en conditions méditerranéennes sur la production de biomasse et la charge de porc noir Majorquin admissible.

Résumé. *Les fourrages ont une importante valeur économique en tant que ressource alimentaire pour le porc noir de Majorque et ils sont largement reconnus pour leurs valeurs environnementales et sociales. La multifonctionnalité de l'élevage est due à son importance économique, environnementale et sociale et est complètement en rapport avec sa durabilité. La production de porc noir de Majorque a été directement liée à l'exploitation de la production agricole. Une meilleure utilisation des fourrages a un net impact financier sur la production de porc noir de Majorque et donc sur sa viabilité. Nous avons étudié la production de biomasse et évalué la charge théorique de porcs noirs de Majorque qu'un pâturage d'orge est capable de supporter, en fonction du nombre d'utilisations. Nous avons utilisé une parcelle expérimentale avec 4 traitements et trois répétitions par traitement. Avec les conditions agronomiques de l'étude, le traitement avec trois utilisations produit plus de biomasse, plus de protéines, plus d'énergie, plus de rations et donc permet une plus forte charge de porc noir de Majorque qu'avec deux ou une seule utilisation.*

Mots-clés. *Porc noir de Majorque – Durabilité – Nombre d'utilisations d'un pâturage.*

I – Introduction

In the Balearic islands forage crops occupy a total of 45749 ha of which 20460 are for feed grains, generally produced under rainfed conditions. The most important and extensive livestock census is the sheep, with over 287000 breeding females (Cifre *et al*., 2007). In second place is the pig sector, in general intensive and with high production inputs, with the exception of the Majorcan black pig production carried out under extensive conditions. The Majorcan Black Pig production system has been directly linked to the exploitation of agricultural production, is linked to grazing (Jaume *et al.*, 2006) and traditionally has been well (Bosch, 1902). Today, forages have significant economic value as a food resource for the Majorcan black pig. The multifunctionality of livestock production is due to its economic, ecological and social importance and is fully linked to its sustainability (Atance *et al.*, 2000). The management of sustainable production systems should be based on the rational use of existing resources on the farm (Roca *et al*., 2008). The use and study of forages to adjust the intensity of pasture is important both to avoid damage to the ecosystem and to profit the resources of grass.

II – Materials and methods

The experimental plot with a crop of barley, destined to be exploited by tooth by livestock, was in a homogeneous area, with a system of continuous blocks with three replications for each treatment. There have been four treatments: one of three cuts in February, April and May (FAM), two of two different cuts in the first court date: February-May (FM) and April-May (AM), and the fourth treatment with a single cut in May. Table 1 shows the abbreviations used to denote treatments.

Table 1. abbreviations used to denote treatments

Treatment	February	April	May
FAM	Cut	Cut	Cut
FM	Cut		Cut
AM		Cut	Cut
M			Cut

At each cutting the following data were recorded: fresh and dry production; on a sample of forage the crude protein content (Kjeldhal method) acid-detergent fiber, neutral-detergent fiber and crude fiber (Van Soest method) were analysed and net energy were estimated. From the data of each cut the cumulative production per hectare of biomass, crude protein and metabolizable energy were calculated. To calculate energy and protein needs the tables proposed by FEDNA (2006) for pigs were used. We used student T-test statistical package Stratgraphics to test the hypothesis that improvement occurs between treatments for a 0.05 significance level.

III – Results

Under the conditions of the present experiment, the effect of the number of cuts has showed significant differences ($p<0.05$) in biomass production, protein and metabolic energy. Thus the treatment with more biomass production and metabolizable energy was that of three cuts (FAM), followed by two cuts (FM, AM) that showed no significant differences between them, and finally single cut treatment (M). The treatment with higher protein production has been three cuts (FAM), followed by two cuts (FM), the two cuts (AM) and finally the single cut (M). The data are presented in Table 2.

It should be noted that the nutritional value of grass, indicated by the relationship crude protein / biomass was increased by increasing the number of cuts. It is not necessary to give the advantage offered by the temporary availability increase in the number of uses.

From a theoretical point of view stocking rate results due to metabolizable energy and crude protein produced by each treatment are presented in Table 3. Grazing three times leads to greater biomass production and consequently, twice as much fattening pigs and sows can graze the same plot as if it is cut once.

Table 2. Accumulated production of biomass, protein and metabolic energy per treatment

Treatment	Biomass (kg/ha)	Protein (kg/ha)	EM (Mcal/ha)
FAM	3.274±750^{a}	528±13^{a}	8.047±465^{a}
FM	2.739±242^{b}	405±34^{b}	6.747±551^{b}
AM	2.894±225^{b}	331±33^{c}	6.714±576^{b}
M	1.863±158^{c}	113±12^{d}	3.546±331^{c}

Table 3. Acceptable stocking rate of Majorcan Black Pig in terms of metabolizable energy and crude protein produced by each treatment

Treatment	Metabolizable Energy		Crude protein	
	Fattening pigs/ha	Sows/ha	Fattening pigs/ha	Sows/ha
FAM	4.1	3.5	5.2	4.8
FM	3.5	3.0	4.0	3.7
AM	3.4	2.9	3.2	3.0
M	1.8	1.6	1.1	1.0

IV – Conclusions

The sustainability of the extensive production of extensive Majorcan black pig is an economic issue. In this regard, using the same inputs, an increase in the number of cuts has obtained higher yields with greater nutritional quality and with a less seasonal annual distribution.

Acknowledgements

The authors thank the pig farm Son Forteza Alaro (Mallorca) cooperation received in this study. The research was funded by the helpline 2009 RAPE.

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Growth of the Krškopolje pig

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Abstract. The Krškopolje pig or blackbelted pig is the only indigenous pig breed in Slovenia. It shows good adaptability to poor feeding and environmental conditions. The breed had an average fertility, a good appetite and high growth ability. The breed is known for its good meat quality, suitable for dried meat products. In our study 40 pigs were included, 22 gilts and 18 barrows. The pigs were kept in one group on bedding (mostly straw) and they were fed with a restricted concentrate and hay at will. At housing, pigs were 140 days old and weighed on average 50 kg. The fixed part of the model for weight included sex, season and age. Age nested within animal and animal was treated as random effect. For daily gain the model included sex and season as fixed part and animal was treated as random effect. At the age 340 days animals weighed an average of 150 kg. Average daily gain during the test ranged between 370 and 780 g, varied depending on the season. Animals, heavier in the beginning, are generally more likely to gain weight, until slaughter. Based on the results we can say that the Krškopolje pigs are suitable for fattening to greater weight.

Keywords. .Pig – Krškopolje pig – Growth – Slovenia.

La croissance du porc Krškopolje

Résumé. *Le porc Kraškopolje est l'unique race de cochon autochtone en Slovénie. Il a démontré une grande adaptabilité à des conditions environnementales et alimentaires pauvres. Cette race se caractérise par une fertilité moyenne ainsi que par un bon appétit et une bonne croissance. La viande de ces cochons est de bonne qualité et se prête bien au produit fabriqué à partir de viande sèche. Dans notre étude, 40 cochons ont été inclus, 22 jeunes femelles (âge <2 ans et n'ayant pas encore été gestantes) et 18 mâles castrés. L'ensemble des porcs constituait un groupe disposant de litière (principalement de la paille) et ils ont été nourris avec un régime restrictif composé d'un mélange de grain (orge, maïs et blé) ainsi que par du foin* ad libitum. *Au moment de la stabulation, les porcs étaient âgés de 140 jours et leur poids moyen était de 50 kg. Concernant notre étude de variation de poids, le sexe, la saison et l'âge ont été considérés comme des variables fixes. L'âge emboîté dans l'animal, ainsi que l'animal, ont été traités en tant qu'effets aléatoires. Pour les études de gain de poids journalier, notre modèle considère le sexe et la saison comme des variables fixes et l'animal comme effet aléatoire. A l'âge de 340 jours, les animaux pesaient 150 kg en moyenne. Le gain de poids par jour, durant l'expérience, oscillait entre 370 et 780 g avec des variations dépendantes de la saison. Aucune différence n'a été notée entre les mâles et les femelles. Les animaux plus lourds au début de l'expérience ont connu une croissance supérieure aux autres et ce jusqu'à abattage. En nous basant sur l'ensemble de ces résultats, nous pouvons conclure que le porc Kraškopolje est approprié pour l'engraissement de par sa bonne augmentation de poids.*

Mots-clés. *Cochon – Porc Kraškopolje – Croissance – Slovénie.*

I – Introduction

The Krškopolje pig is the only preserved indigenous pig breed in Slovenia. The Slovenian indigenius pig is black, with white unbroken belt over the shoulders and down to both front feet (Fig. 1). Under extensive management, it shows good adaptability to pooor fedding and breeding conditions (Šalehar, 1994). The breed was formed and developed under the influence of the environment, animal husbandry, selection, and domestic economic conditions. The breed has an average fertility, relatively high losses, a good apetite and growth ability. Adult animals weigh 250-300 kg.

Fig. 1. The Krškopolje pig (Foto: Š. Malovrh).

Between 1970 and 1990, the Krškopolje breed was more than ever, left to itself. During this time, there wasn’t any systematic breeding work. There were some uncontrolled integration of other breeds used for mating due to low number of sires. In 1990 the breed reconstruction began by increasing the population size and setting up a breeding program (Šalehar *et al*., 1992). German Saddleback was included in 2003 in order to reduce the high risk of inbreeding (Šalehar, 2008).

The high amount of intramuscular fat, marbling and darker color have been highlitghed as some of the most reletavant quality aspectc in muscles from Krškopolje pigs (Eiselt and Ferjan, 1972; Čandek-Potokar *et al*., 2003; Furman *et al*., 2010). Because of these properties the meat of Krškopolje pigs is suitable for processing into dry-cured products. Krškopolje dry-cured products are valuable Slovenian traditional meat products. Unfortunately, the Krškopolje pigs cannot be rared intensively because they become too fatty (Furman *et al*., 2010).

The objective of this study was to examine the growth of the Krškopolje pigs breed.

II – Materials and methods

In the experiment, 40 Krškopolje pigs were included, 18 barrows and 22 gilts. All animals were raised on the same farm. The pigs were kept in one group on bedding (mostly straw) and they were fed with a restricted concentrate (wheat, barley, maize with vitamin mineral supplement) and hay at will. Animals were given water in the sink, which was cleaned twice daily and filled with fresh water. The area of pen was 150 m^2, which means 3.75 m^2 per animal. Barn, where pigs have been established, had a foreign climate.

At housing, pigs were 138.5 days old and weighed on average 48.9 kg (Table 1). The gilts were on average slightly lighter and for 2.9 days younger than barrows. At slaughter the average body weight of animals was 139 kg at the age 297.8 days. At the time of slaughter gilts were on average 4 kg heavier.

Table 1. Average age and weight with standard deviation for the pigs at the beginning of the experiment and before slaughter

	Beginning of the experiment		End of the experiment	
	Age (day)	**Body weight (kg)**	**Age (day)**	**Body weight (kg)**
Together	138.5 ± 11.8	48.9 ± 7.5	297.8 ± 33.1	139.0 ± 16.4
Barrows	136.9 ± 12.9	49.2 ± 8.3	288.3 ± 33.5	136.9 ± 17.2
Gilts	139.8 ± 11.0	48.8 ± 6.9	305.5 ± 31.5	140.7 ± 16.0

The fixed part of the model for weight included gender (G_i), season (S_J) and age (equation 1). Age nested within animal and animal was treated as random effect.

$$y_{ijk} = \mu + G_i + S_j + b_{ij}(x_{ijk} + \bar{x}) + a_{ijk} + e_{ijk} \qquad (1)$$

For daily gain the model included gender and season as fixed part and animal was treated as random effect (equation 2).

$$y_{ijk} = \mu + G_i + S_j + a_{ijk} + e_{ijk} \qquad (2)$$

In matrix notation, Eq. 1 and Eq. 2 can be written as: $\mathbf{y} = \mathbf{X}\beta + \mathbf{Z}_a a + \mathbf{e}$

where the vectors **y, a** and **e** are assumed to have the following structure of the expected value and variance:

$$E\begin{bmatrix}\mathbf{y}\\ \mathbf{a}\\ \mathbf{e}\end{bmatrix} = \begin{bmatrix}\mathbf{X}\beta\\ \mathbf{0}\\ \mathbf{0}\end{bmatrix}; V = \begin{bmatrix}\mathbf{a}\\ \mathbf{e}\end{bmatrix} = \begin{bmatrix}\mathbf{I}_a\sigma_a^2 & \mathbf{0}\\ \mathbf{0} & \mathbf{I}_e\sigma_e^2\end{bmatrix}$$

Data were processed in the SAS statistical package (SAS Inst. Inc., 2001). Restricted likelihood method (REML) was used for linear mixed model.

III – Results

Season, age and animal were statistically significant in the model for body weight (Table 2). Age was the model for body weight included as a linear regression. Only season was proved as statistically significant as the model for the average daily gain. Animal and gender were not statistically significant.

Table 2. P-values of effect in model for body weight and daily gain

Effect	Body weight	Daily gain
Gender	0.5286	0.4767
Season	<0.0001	<0.0001
Age	<0.0001	/
Animal	<0.0001	0.1821

The biggest difference in body weight between the sexes at the age of 300 days, but differences were statistically insignificant, however.

Statistically significant effect of animals tells us that the animals had increased body weight at the start, usually before reaching a body weight that we want to slaughter (Fig. 2). At the age of about 140 days the average weight of animals was 50 kg and 150 kg at the age 340 days.

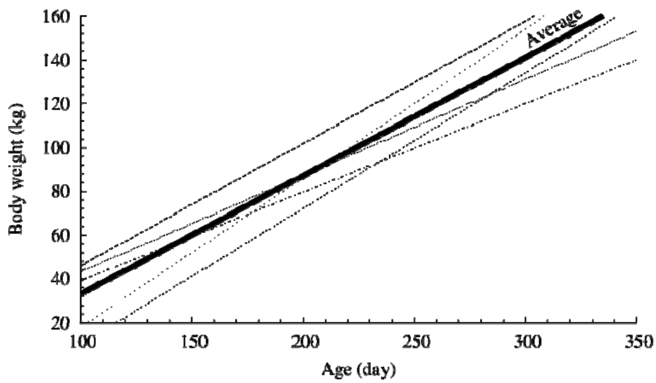

Fig. 2. Growth curve for individual animals.

In the literature, we identified records experiments where Krškopolje pigs fattened at lower body weight than we fattened pigs in our experiment. In one experiment, pre-slaughter pigs weighing between 110 and 167 kg (Kastelic, 2001). Otherwise, the weight at slaughter was between 107.8 kg (Ferjan, 1969) and 117.7 kg (Eiselt and Ferjan, 1972).

Daily gain was highest at around 60 kg body weight. During this period, the average daily gain was 727 (gilts) and 781 g per day (barrows). A smaller daily gains, below 400 g per day, were at about 100 kg body weight. During this period the temperature was below freezing barn so the pigs needed more energy to maintain body temperature.

Solid daily gain (over 550 g/day) was also at 150 kg body weight. Daily gain varies between season, which in this case represents the sequence weighting. The seasonal variations is presented - as an exception to the rest of the first season (Fig. 3). Calculated daily gain was not comparable with the Krškopolje breed pigs which were in the experiment growth rate of 39 to 132 kg weight .In this case, the average increment was 1003 g per day.

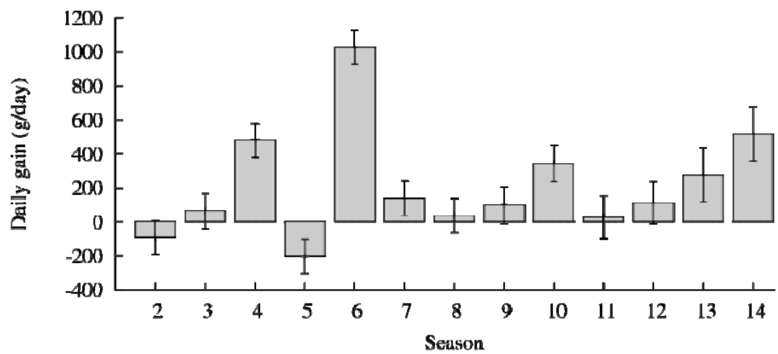

Fig. 3. Daily gain, depending on the season.

Daily gain of the Krškopolje pigs in our experiment can be compared with the increment in the experiment that was carried out by Krhin (1959), where average daily gain was 550 g per day. The average daily gain of the Krškopolje pigs was estimated at the 461 g per day (Ferjan, 1969) and 432 g per day (Eiselt and Ferjan, 1972). In both cases, the pigs increment was worse than in our experiment.

IV – Conclusions

Indigenous breeds have become an integral part of biodiversity, which makes the Krškopolje pig, as the only Slovenian indigenous pig breed, to gain in importance. There are little data on slaughter and fattening characteristics for this breed. Body mass varied both between animals and by age. There were no statistically significant differences between gender. All animals at slaughter exceeded 120 kg. In our experiment, daily gain of the Krškopolje pig was on average 557 g. Based on the results of the experiment it can be argued that the observed Krškopolje pigs are suitable for fattening at the greater weight.

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Effect of dietary fat and restriction on carcass and meat quality of Iberian pigs

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Abstract. A total of 160 Iberian x Duroc pigs (50% males and 50% females, all castrated) of 40.3±2.7 kg of initial BW was used to study the influence of dietary fat source and feed restriction on carcass and meat quality. There were three experimental periods: (i) grower: 40-80 kg BW, 98 d of trial, with a restricted common diet; (ii) fattening: 80-114 kg BW, four treatments arranged as a 2 x 2 factorial design: *ad libitum* (31d) *vs* restricted (51d) with diets containing Iberian pig lard –IL– *vs* oleic acid enriched sunflower oil –OESO–, and (iii) finisher: 114 kg slaughter, 77d, diets based on IL or OESO *ad libitum*. Total experimental lengths were 206 and 226 d, for pigs fed *ad libitum* and restricted, respectively. At the end of the trial, restricted pigs showed lower backfat depth than pigs fed *ad libitum*. However, pigs restricted in the grower period had better carcass yield than *ad libitum* pigs but no significant differences were found for ham, shoulder and loin yield. In addition, feed restriction decreased L* and b* values of the loin. Finally, the fatty acid profile of the intramuscular fat was not affected by fed restriction and fat source.

Keywords. Iberian pigs – Fat sources – Feed restriction.

Effet des graisses alimentaires et de la restriction sur la carcasse et la qualité des viandes de porc Ibérique

Résumé. *Un total de 160 porcs croisés Ibérique x Duroc (50% mâles et 50% femelles, tous castrés) de 40,3 ± 2,7 kg de poids vif initial ont été utilisés pour étudier l'influence de l'origine des graisses alimentaires et du rationnement sur la carcasse et la qualité de la viande. L'essai a été partagé en trois périodes expérimentales : (i) croissance: 40 à 80 kg de PV, durée de 98 jours, alimentation commune* ad libitum *; (ii) engraissement : 80 à 114 kg de PV, comparaison d'un régime alimentaire contenant de la graisse de porc Ibérique (MAN) vs un régime contenant de l'huile de tournesol riche en acide oléique (GRO), distribués soit* ad libitum *pendant 31 jours soit en rationnement pendant 51 jours ; et (iii) finition: 114 kg de PV à l'abattage, 77 jours, régimes à base de MAN ou de GRO, dans les deux cas,* ad libitum*. La durée totale de l'essai a été de 206 et 226 jours respectivement pour les porcs nourris* ad libitum *et ceux rationnés. À la fin de l'essai, les porcs rationnés ont présenté une épaisseur de gras dorsal inférieure à celle des porcs* ad libitum*. Cependant, les porcs rationnés ont eu un rendement de carcasse supérieur à celui des porcs* ad libitum*, mais aucune différence significative n'a été trouvée pour le jambon, l'épaule et la longe. En outre, le rationnement alimentaire a diminué les valeurs L* et b* de la longe. Enfin, le profil de la graisse intramusculaire en acides gras n'a pas été affecté par le rationnement alimentaire ni par la source de matières grasses.*

Mots-clés. *Porcs Ibériques – Graisses alimentaires – Rationnement alimentaire.*

I – Introduction

Under the current legislation of Spain regarding to Iberian pig production, Duroc sires can be used in the production of Iberian (IB) pigs to improve reproductive performance and feed efficiency, but these pigs must be slaughtered at ages of 10 months or older (Boletín Oficial del Estado, 2007). When IB x Duroc pigs are provided *ad libitum* (AL) access to their feed, they reach excessive BW at that age, with economic disadvantages because of increased fat content and excessive size of the primal cuts. In this sense, one strategy to reduce the growth of animals is the feed restriction. The feeding program used in most farms consists in a restricted feeding during growing period for developing of the bones and improve feed efficiency, and subsequently, *ad libitum* access to a high caloric diet rich in oleic acid to take advantage of compensatory growth until slaughter. However, the restricted feed intake (FI) level may affect the fatty acid (FA) deposition because the endogenous lipid synthesis is reduced (Nürnberg *et al.,* 1998), and compensatory growth could affect carcass and primal cuts yield.

On the other hand, feed is probably the most influential factor on the final quality of meat, since there is a close relationship between feed intake of animal and body composition (Ruiz *et al.,* 2002). Traditionally it was considered that the feed received at the final stage of fattening determined the FA profile of animal fat (Cava *et al.,* 1999). However, that FA profile could be conditioned by feeding during the growing period (López Bote *et al*., 1999 and 2000), and could be an interaction between feed restriction and fat source. Therefore, the aim of the present study was to evaluate the influence of the feed restriction and the fat source on carcass and meat quality of Iberian pigs destined to dry-cured product industry.

II – Materials and methods

1. Experimental design and animals

A total of 160 Iberian x Duroc pigs (50% males and 50% females, all castrated) of 40.3 ± 2.7 kg of initial BW, and 147 d of age was used to study the influence of dietary fat source and feed restriction on carcass and meat quality. Pigs were managed from birth to initiation of the experiment according to standard commercial procedures. Prior the beginning of the trial, animals were allotted in 16 blocks (10 pigs per block) of 250 m^2 according to their BW and sex in order to obtain homogeneous replicates. There were three experimental periods (Table 1): (i) grower: 40-80 kg BW, 98 d of trial, with a restricted common diet; (ii) fattening: 80-114 kg BW, four treatments arranged as a 2 x 2 factorial design: *ad libitum* (AL, 31d) *vs* restricted (FR, 51d) with diets containing Iberian pig lard –IL– *vs* oleic acid enriched sunflower oil –OESO; and (iii) finisher: 114 kg–slaughter, 77 d, diets based on IL or OESO *ad libitum*. Feed restriction was 3% BW of pigs, recalculating the amount of feed every 15 d. With the aim of reach similar final BW between all pigs, the total experimental lengths were 206 and 226 d, for pigs fed *ad libitum* and restricted, respectively.

Table 1. Experimental design of the trial.

Treatment	Fattening (80-114 kg BW)	Finisher (114 kg BW-slaughter)
FR-IL	Restricted -IL-	*Ad libitum* -IL-
AL-IL	*Ad libitum* -IL-	*Ad libitum* -IL-
FR-OESO	Restricted -OESO-	*Ad libitum* -OESO-
AL-OESO	*Ad libitum* -OESO-	*Ad libitum* -OESO-

Grower (40-80kg BW): Restricted -IL-.

2. Experimental diets

All diets were formulated by SAT Villa Vieja and Imasde Agroalimentaria, S.L. and met or exceeded the nutrient requirements of pigs (NRC, 1998) for Iberian pigs. All diets were calculated to be isonutritive except for net energy, due to the replacement of IL by OESO. The composition and the estimated (FEDNA, 2003) or determined (AOAC, 2000) nutrient values of the diets are shown in Table 2. Diets were manufactured by SAT Villa Vieja and were offered in mash form.

3. Carcass traits

At the preplanned slaughter BW (with 206 and 226 d of trial for *ad libitum* and restricted pigs, respectively) all pigs were weighed and identified individually and transported to a commercial abattoir (Jamón y Salud, S.A., Badajoz, Spain) and were stunned with CO_2, exsanguinated and eviscerated. Then all carcasses with the head were weighed individually to obtain carcass yield and backfat (BF) thickness was measured between the third and fourth last ribs on the middle of carcass (skin included).

Table 2. Experimental diets

	40-80 kg BW	80-114 kg BW		114 kg BW-slaughter	
	Common diet	**IL**	**OESO**	**IL**	**OESO**
Ingredients %					
Barley, wheat and corn	71.70	70.10	70.10	77.10	77.10
Soybean meal, 44% CP	20.20	13.90	13.90	11.00	11.00
Wheat bran	4.00	7.80	7.80	4.00	4.00
Iberian lard	1.00	5.00	-	5.00	-
Sunflower meal	-	-	5.00	-	5.00
Calcium carbonate	0.80	0.95	0.95	1.20	1.20
Dicalcium phosphate	1.30	1.25	1.25	1.10	1.10
Sodium chloride	0.40	0.40	0.40	0.40	0.40
L-Lysine 78,8%	0.10	0.10	0.10	-	-
V&M premix	0.50	0.50	0.50	0.20	0.20
Calculated nutrient composition[a], %					
NE, kcal/kg	2.255	2.430	2.450	2.479	2.499
Crude protein, %	17.1	15.3	15.3	14.1	14.1
Lysine, %	0.89	0.77	0.77	0.61	0.61
Ether extract, %	3.1	6.8	6.8	6.8	6.8
Total ash, %	4.7	5.0	5.0	4.9	4.9
Ca, %	0.72	0.76	0.76	0.81	0.81
P Total, %	0.64	0.76	0.63	0.58	0.58
C16:0, %	0.49	1.43	0.52	1.42	0.52
C18:0, %	0.15	0.67	0.21	0.67	0.21
C18:1, %	0.74	2.32	4.10	2.31	4.09
C18:2, %	0.99	1.25	1.09	1.23	1.07
C18:3, %	0.08	0.13	0.09	0.12	0.08
Analyzed composition[b], %					
Crude protein, %	16.2	15.1	14.8	13.4	13.6
Crude fiber, %	4.8	4.9	4.7	4.7	4.7
Ether extract, %	3.1	4.3	4.1	6.0	5.6
Total ash, %	4.5	4.4	4.7	4.6	4.9

[a]According to FEDNA (2003).[b]According to AOAC (2000).

Before quartering carcasses, all hams, shoulders and loins were identified individually. Afterward, hams, shoulders and loins were trimmed of external fat and weighed in order to calculate the yield of primal cuts.

4. Meat quality traits and FA profile

Thirty-two carcasses (sixteen from each treatment, two per block) were randomly selected and *Longissimus dorsi* muscle (LM) samples were taken at the level of the last rib. Meat samples were stored in individual plastic bags and transported to Universidad de Extremadura for subsequent analyses. Instrumental colour (L*, a*, and b*) was determined on one chop of 1 cm of thickness obtained from each sample after a 30 min bloom period. Colour was measured three times per sample using a Minolta Chromameter CR-300 (Minolta Camera Corp., Meter Division, Ramsey, NJ).

Protein content of each sample was determined using the methodology described by Lowry *et al.* (1951). Total lipids of loin samples were extracted with chloroform:methanol (2:1 v/v) according to the method of Folch *et al.* (1957). After solvent evaporation under nitrogen, the neutral lipid (NL) and the polar lipid (PL) fractions were isolated according to the method of Ruiz et al. (2004) using NH2-aminopropyl cartridges. Fatty acid methyl esters (FAMEs) from lipid fractions were obtained by acidic transesterification, following the method described by Sandler and Karo (1992). FAMEs were analysed by gas chromatography, using a Hewlett-Packard HP-5890A gas chromatograph, equipped with an on-column injector and a flame ionisation detector (FID). Individual compounds were identified by comparing their retention times with those of standards (Sigma, St. Louis, MO).

5. Statistical analysis

The individual pig was the experimental unit for analysis of all data. Data were analyzed as a completely randomized design with four treatments in a 2 x 2 factorial arrangement using the GLM procedure of SAS (SAS Inst., Inc., Cary, NC, USA). The model included the feeding regimen (AL *vs* FR), fat source (IL *vs* OESO), and their interactions. Sex of animals was included as fixed effect.

III – Results and discussion

Pigs restricted were slaughtered with 171.5 kg and 226 d of trial (373 d of age), whereas AL pigs were slaughtered with 167.3 kg and 206 d of trial (353 d of age). The effects of feeding program and fat source of the diet on backfat thickness at the level of P2 point and carcass and primal cuts yield are presented in Table 3.

Table 3. Effect of feeding regimen and fat source on backfat (BF) thickness and carcass and primal cuts yields

	BF thickness, mm	Carcass and primal cuts yield			
		Carcass, %	Ham, %	Shoulder, %	Loin, %
Feeding regime					
AL	7.50	79.99	17.17	11.26	3.51
FR	7.08	80.67	17.16	11.29	3.50
Fat source					
OESO	7.25	80.38	17.16	11.25	3.52
IL	7.34	80.28	17.16	11.30	3.49
SEM[1] (n=75)	0.10	0.15	0.11	0.07	0.05
P-value[2]					
Feeding	0.005	0.002	NS	NS	NS
Fat	NS	NS	NS	NS	NS

[1]SEM=Standard error of the mean; [2]Interactions were not significant.

At the end of the trial, restricted pigs showed lower backfat depth than pigs fed *ad libitum* (P<0.01). This result is in agreement with those observed by Daza *et al.* (2007) and Serrano *et al.* (2009). However, in the grower period pigs restricted had better carcass yield than *ad libitum* pigs (P<0.01) but no significant differences were found for ham, shoulder and loin yield. Also, no differences were found between fat sources of the diet for backfat thickness and carcass and primal cuts yield of the IB x Duroc pigs.

The effects of feeding program and fat source of the diet on instrumental colour and chemical composition of the meat are presented in Table 4. Feed restriction decreased L* and b* values of the loin (P<0.01). However, chemical composition of the meat was not affected by the feeding regimen of the animals.

Pigs fed OESO tended to show lower b* value than pigs fed IL (P=0.08). Also, pigs fed OESO showed less protein and fat content on *Longissimus dorsi* than pigs fed IL (P≤0.05).

Table 4. Effect of feeding regimen and fat source on instrumental colour and chemical composition of meat.

	Instrumental colour			**Chemical composition**		
	L*	**a***	**b***	**Moisture %**	**CP %**	**Fat %**
Feeding regime						
AL	50.55	12.78	8.76	68.21	18.74	4.00
FR	46.12	13.96	5.17	69.33	19.38	3.09
Fat source						
OESO	48.17	13.38	6.48	69.00	18.08	2.92
IL	48.50	13.36	7.45	68.53	20.05	4.16
SEM[1] (n=16)	0.92	0.50	0.36	0.59	0.66	0.37
P-value[2]						
Feeding	0.003	NS	<0.001	NS	NS	NS
Fat	NS	NS	*0.076*	NS	0.050	0.029

[1]SEM=Standard error of the mean. [2]Interactions were not significant.

The effects of feeding program and fat source of the diet on fatty acid profile of the neutral and polar fractions of the intramuscular fat of meat are presented in Table 5. Feed restriction tended to decrease the proportion of linolenic acid (C18:3) on the neutral fraction, and the palmitic acid (C16:0) of the polar fraction regarding *ad libitum* feeding (P=0.06). However, feeding regimen did not affect significantly the remaining FA studied of the neutral and polar fractions of the intramuscular fat.

Pigs fed OESO showed a higher proportion of linolenic acid on the neutral fraction (C18:3) than pigs fed IL (P<0.05), but no significant differences were found for the remaining FA of the neutral fraction of intramuscular fat. Also, the inclusion of OESO in the diets increase the proportion of stearic acid (C16:0), and tended to show higher proportion of oleic acid (C18:1) of the polar fraction than diets with IL (P<0.05, and P=0.06, respectively).

Also, a feeding regimen x fat source interaction was found for the proportion of linolenic acid of the neutral fraction of the intramuscular fat (P<0.05). In this sense, while the fat source of the diet did not affect the linolenic acid (C18:3) content in restricted pigs, pigs fed *ad libitum* consuming the IL diet showed greater linolenic acid proportion than pigs fed OESO.

Table 5. Effect of feeding regimen and fat source on fatty acid profile of the neutral and polar fractions of the intramuscular fat of meat

Fatty acid, %	Feeding regimen		Fat source		SEM[1] (n=16)	P-value		
	AL	FR	OESO	IL		Feeding	Fat	Feeding x Fat
Neutral lipids								
Palmitic (C16:0)	24.73	25.14	24.84	25.03	0.23	NS	NS	NS
Palmitoleic (C16:1)	4.46	4.52	4.38	4.61	0.10	NS	NS	*0.077*
Stearic (C18:0)	11.87	12.31	12.18	12.00	0.33	NS	NS	NS
Oleic (C18:1)	53.27	52.49	52.95	52.81	0.37	NS	NS	NS
Linoleic (C18:2)	2.68	2.49	2.65	2.51	0.12	NS	NS	*0.051*
Linolenic (C18:3)	0.16	0.14	0.16	0.13	0.01	*0.063*	0.011	0.023
Polar lipids								
Palmitic (C16:0)	23.78	22.03	22.82	22.99	0.61	*0.057*	NS	NS
Palmitoleic (C16:1)	1.40	1.32	1.30	1.42	0.09	NS	NS	NS
Stearic (C18:0)	9.20	8.68	8.40	9.48	0.33	NS	0.038	NS
Oleic (C18:1)	24.38	23.55	25.11	22.81	0.79	NS	*0.061*	NS
Linoleic (C18:2)	33.87	34.70	34.85	33.72	1.14	NS	NS	NS
Linolenic (C18:3)	0.42	0.48	0.39	0.51	0.06	NS	NS	NS

[1]SEM=Standard error of the mean.

IV – Conclusions

A feed restriction of 3% BW of pigs from 40 to 80 kg is enough to obtain commercial slaughter weights at 10 months of age in IB x Duroc pigs. Moreover, an additional feed restriction until 114 kg BW decrease backfat thickness, improve carcass yield, and modify the instrumental colour of the meat, without affecting primal cuts yield, chemical composition, and FA profile of the meat. However, the whole fattening-finishing period was increased in 20 d. As we expected, the fat source of the diet did not affect backfat depth, carcass traits or instrumental colour of meat, and results in a slight differences in the FA profile of the intramuscular fat of the meat.

Acknowledgments

The research was supported by Ministerio de Industria, Turismo y Comercio (Project CDTI 2004-0137), and Junta de Extremadura (Project AI 0201031) of Spain.

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Preliminary results of productivity of Gochu Asturcelta's breed in semi-extensive systems

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Abstract. The aim of this work was to carry out preliminary studies of productivity of Gochu Asturcelta´s breed in semiextensive systems. There were confirmed two different strategies of growing and fatteninig of an autochthonous endangered pig breed, following the recommendations given for Iberian pig. A total of 19 animals in 2008 and 22 in 2009 were tested. The first strategy used rearing feed *ad libitum* from the weaning, except a restriction to 2.5 kg $pig^{-1}day^{-1}$ in the last month. In other one, the rearing feed was replaced to five months postwean for other feed: growing *ad libitum* (from 5 to 8 months) and fattening to dose of 2-2.5 kg $pig^{-1}day^{-1}$. The finishing in both years was done with different proportions of non-transgenic cereals. There were no differences in the live weight curves of evolution in both cases, turning out to be a better feed conversion rate with the second strategy (5.1 *vs* 5.3). The results showed that it is interesting to shorten the initial period of supply *ad libitum* and to restrict later to 2-2.5 kg $pig^{-1}day^{-1}$, using feed mixtures with 3200 kcal of digestible energy/kg dry matter and 10-12% crude protein, similar to those of the Iberian pork. However, further work is needed to investigate the effects of feeding levels on growth and meat quality of Gochu Asturcelta pig.

Keywords. Celtic trunk – Local pig – Semi-extensive production – Management.

Résultats préliminaires de productivité de la race Gochu Asturcelta en systèmes semi-extensifs

Résumé. *On a contrasté deux différentes stratégies de croissance-engraissement de porcs de race autochtone Gochu Asturcelta en régime semi-extensif, en suivant les recommandations données pour le porc Ibérique. Dans le premier cas, on a utilisé exclusivement un aliment pour élevage* ad libitum *depuis le sevrage, sauf restriction à 2,5 kg/porc/jour dans le dernier mois. Dans l'autre, on a remplacé l'aliment d'élevage à cinq mois post-sevrage par d'autres aliments de croissance* ad libitum *(5 à 8 mois) et d´engraissement à raison de 2-2,5 kg/porc/jour (> 8 mois). Il n'y a pas eu de différences dans les courbes d'évolution du poids vif dans les deux cas, un meilleur indice de conversion étant obtenu avec la seconde stratégie (5,1 vs 5,3). Il est donc intéressant de raccourcir la période initiale d'alimentation* ad libitum *et de restreindre ensuite à 2-2,5 kg/porc/jour, en utilisant les recommandations pour croissance et engraissement du porc Ibérique (3200 kcal d'énergie digestible/kg de matière sèche et 10-12% de protéines brutes). Toutefois, des travaux supplémentaires sont nécessaires pour étudier les effets de ces niveaux d'alimentation sur la croissance et la qualité de la viande de porc Gochu Asturcelta pour déterminer le niveau optimal d'alimentation.*

Mots-clés. *Tronc celtique – Race locale – Production semi-extensive – Gestion.*

I – Introduction

The presence of swine breeding in Asturias (*Sus célticus)* finds its origin in the pre-Roman period (V century BC), (Álvarez Sevilla, 2001). In the 20th century socio-economic transformations in the agro-food system, intensification and industrialisation, variations in land use and massive utilisation of commercial breeds determined the decline of local breeds (Franci *et al., 2007)*. The National Register of this local breed was in 2007 (BOE no. 21 ; 24/01/2007) by the Pig Breeders Association (ACGA) at the request of the Spanish Ministry for Agriculture and Fisheries. According to Royo *et al.* (2007), autochthonous Gochu Asturcelta porcine race is genetically close to the current white pigs of Northern-Central Europe and diverge significantly at nuclear loci with respect the Iberian pig. Nevertheless, the Celtic and Iberian pigs have in common their wildness. The above mentioned suggests that also it will present nutritive requirements more similar to those of the Iberian pork, which quantification was and continues being the aim of several researches. The adoption of recommendations for gestation, lactation and rearing feeds destined to Iberian pig was decisive in the process of recovery of the Gochu Asturcelta breed (López Bote *et al.,* 2000), in the returning to local pig production in Asturias and they were the crucial way to obtain optimal results, although their percentage of registration is still very low. The production structure is constituted by a very small number of traditional farmers that sell on farm their production. In this situation, it is necessary to confirm the effects of feeding and rearing with nutritional requirements for Iberian pork with the "montanera, recebo and cebo" systems as a model (Barea *et al.*, 2007; Nieto *et al*., 2009) on growth performance. The aim of this work was to carry out preliminary studies of productivity of Gochu Asturcelta´s breed in semi-extensive systems, following the recommendations given for Iberian pig and return to regional customs of Asturias.

II – Materials and methods

For two successive years, 2008 and 2009, different treatments of growth and fattening were established with animals obtained in the own core of multiplication of the SERIDA. The animals were proceeding in both cases of near births, which were allowing the adoption of an average to which recounting the later ages. In 2008, a total of 19 pigs were used (9 castrated males plus 10 females) and in 2009 a total of 22 animals were employed (11 castrated males plus 11 females). The weaning took place at 46 days in 2008 and at 57 days in 2009, with respective mean values of weight of 12.35 and 15.89 kg. Table 1 shows the different durations of growth and fattening periods during 2008 and 2009, as well as the doses of feed corresponding to each one. The principal difference takes root in that during 2009 the phase of growth with feed *ad libitum* was slow and the duration of fattening with restricted supply was increased. Additionally, in 2008 rearing feed was exclusively used, whereas, in 2009, it was replaced immediately with one ratio of growth and other one of fattening. Following the rules of the ACGA the feeding system for finishing period in both years was done with different proportions of non-transgenic cereals.

Table 1. Different targets of growth, fattening and finishing on 2008 and 2009 years

Period	Year 2008			Year 2009		
	Age interval (months)	Feed	Dose (kg pig^{-1}day^{-1})	Age interval (months)	Feed	Dose (kg pig^{-1}day^{-1})
Growing	1.5 - 7.5	Rearing	*Ad libitum*	1.5 - 6	Rearing	*Ad libitum*
Fattening	7.5 - 11.5	Rearing	2.5	6 - 10	Rearing	2
				10 - 12.5	Fattening	2.5
				vs		
				10 - 14.5	Fattening	2.5
Finishing	11.5 - 16 vs 18	Fatteninig	*Ad libitum*	12.5 - 14	Fattening	*Ad libitum*
				vs		
				14.5 - 16	Fattening	*Ad libitum*

The respective nutrient contents and energetic value estimation (EvaPig® software, 2008; Noblet *et al.,* 2004) can be seen in Table 2. The live weight was controlled to regular intervals of four weeks, as well as the consumption of feed when it was supplied *ad libitum*. A semiextensive regime was always followed, that is, supply on the basis of fodder but in outdoor parcels with spontaneous vegetation, camping type refuges and free space to favor the natural behaviour of the animals. The health and behaving of pigs were regularly inspected.

Table 2. Chemical composition (% on DM) and estimated digestible energy (DEf) of different feed mixtures used in the years 2008 and 2009

Year	2008 and 2009	2009	2009
Feed	**Rearing**	**Fattening**	**Finishing**
Crude protein	17.4	12.0	9.3
Ether e xtract	4.4	3.4	3.0
Crude fiber	6.6	3.5	3.1
DE_f (MJ $kgDM^{-1}$)	13.9	13.5	13.4

III – Results and discussion

All pigs remained in good health throughout the experimental periods and no feed refusals were detected.

Figure 1 shows the evolution of the live weight of the animals with both different strategies of growth and fattening applied throughout 2008 and 2009 years. The figure already shows a close up view of the feed conversion rates (kg of feed kg^{-1} of increase of weight) and daily increase of weight (kg) (see Table 3). During the period from five to six months, with feed ingestion *ad libitum*, there were no differences in conversion rates and average daily gain (ADG) between rearing and growth feed mixtures. From eight to 10 months, employing fattening feed, which has less total energetic and protein contents that rearing feed and has minor total consumption too, it was obtained the same final live weight for both feed mixtures. The methodical pre-established for control of the evolution of the live weight was identical for both years and it was determined to 10 months. It turns out to be, so, the most appropriate age to effect productive contrasts.

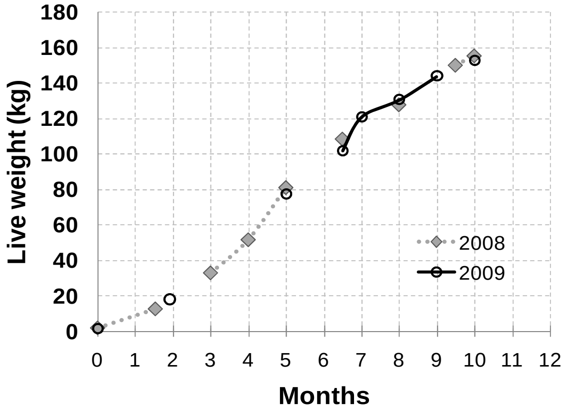

Fig. 1. Monthly live weight evolution along 2008 and 2009 years.

As expect, the final average live weight of the males turned out to be superior to that of the females (162.3 *vs* 143.5 kg; $p<0.01$). However, as for the different strategies, both males and females considered together in results, there are no significant differences, (155.2 kg in 2008 vs

152.2 kg in 2009). Nevertheless, the total consumption of feed up to 10 months of age is different: 727.5 kg in 2008 *vs* 577.5 kg in 2009 (P<0.001). According to the mentioned values, the global feed conversion ratio from the weaning up to 10 months would be of 5.1 in 2008 *vs* 5.3 in 2009 and the results concerning the same period for ADG were 593 g for 2008 *vs* 509 g for 2009 respectively. The results are in agreement with those of Barea *et al*. (2007) and Nieto *et al*. (2009) report on Iberian pig.

In addition to the advantage of using more economical feed for equal deposition of protein in the body of the animals, we have to mention the environmental importance of a minor excretion of urinary N to the environment.

Table 3. Feed conversion rates (kg feed kg live weight gain^{-1}) and average daily gain (ADG, kg) according to the different target feeding used in 2008 and 2009

		Year 2008		
Period	**Feed**	**Dose**	**Feed: Gain**	**ADG**
Weaning-3 months	Rearing	*Ad libitum*	2.81	0.534
3-4 months	Rearing	*Ad libitum*	4.21	0.670
4-5 months	Rearing	*Ad libitum*	4.09	0.727
5-6 months	Rearing	*Ad libitum*	4.78	0.643
6,5-8 months	Rearing	*Ad libitum*	8.09	0.544
8-9,5 months	Rearing	*Ad libitum*	8.50	0.294
9.5-10 months	Rearing	2.5 kg pig^{-1} day^{-1}	3.38	0.740
		Year 2009		
Period	**Feed**	**Dose**	**Feed: Gain**	**ADG**
Weanig-5 months	Rearing	*Ad libitum*	4.20	0.672
5-6 months	Growing	*Ad libitum*	4.77	0.684
6-7 months	Growing	*Ad libitum*	3.46	0.578
7-8 months	Fattening	2 kg pig^{-1} day^{-1}	5.14	0.389
8-9 months	Fattening	2 kg pig^{-1} day^{-1}	5.58	0.448
9-10 months	Fattening	2.5 kg pig^{-1} day^{-1}	8.85	0.282

The need for more investigations on growth remains open and the objectives should be considered because no clear trends in the market can be detected today.

IV – Conclusions

The present results in semi-extensive farming of the Gochu Asturcelta pigs suggest that, after the weaning, it is possible to shorten the period of supply *ad libitum* during growth performance. During the following phase of fattening, it is interesting to restrict both energetic and protein supplies, dosing 2-2.5 kg feed pig^{-1}day^{-1} with 13.2 MJ kg DM^{-1} of digestible energy and 12-10% of crude protein on DM basis. However, further work is needed to investigate the effects of those feeding levels on growth and meat quality of Gochu Asturcelta pig to determine the optimal feeding level.

Acknowledgements

The authors are grateful to the field support staff and to the laboratory of Nutrition staff of the Nutrition, Grasslands and Forages Department of SERIDA for the well done tasks. The investigation has been supported by the Regional Ministry of Rural Environmental and Fisheries of the Principality of Asturias and SERIDA.

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Physicochemical characterization of Majorcan Black Pig lard

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Abstract. The *Ensaimada de Mallorca* is a typical sweet from the Island, made of flour and animal fat, and it is covered by a Protected Geographical Indication. Its name derives from *saïm*, which means pork lard in Catalan, and until the first half of the twentieth century it came exclusively from Majorcan Black Pigs. Currently, the Majorcan Black Pig lard is not used any more in the elaboration of *Ensaimada*. In the present study the physicochemical properties and thermal behaviour of fat from Majorcan Black Pigs were determined. The effect of the different origins of fat tissue from the pig carcass (flare fat *vs* intermuscular fat) on the elaboration process and quality of *Ensaimada* was assessed, using a reference scale set up by the Regulatory council of the *Ensaimada de Mallorca*. The results obtained showed that the physicochemical characteristics and behaviour of fat from Majorcan Black Pig were dependent on the type of fat tissue used in its preparation. The Majorcan Black Pig lard showed a monounsaturated fatty acid composition of 45.6% ± 2.6 in flare fat, and 50.9% ± 3.8 in intermuscular fat, due mainly to high oleic acid content. The type of fat that showed more similar characteristics and behaviours to those defined as the standard was the flare fat.

Key words. Majorcan black pig – *Ensaimada*.

Caractérisation physico-chimique du saindoux de porc noir de Majorque

Résumé. *L'*Ensaimada de Mallorca *est un gâteau typique de l'île à base de farine et de saindoux, qui est couvert par une Indication Géographique Protégée. Son nom dérive du mot saïm, ce qui en majorquin signifie saindoux. À Majorque le saindoux, jusqu'à la première moitié du XX^{e} siècle, provient exclusivement du Porc Noir de Majorque. Actuellement, le saindoux du Porc Noir de Majorque n'est pas utilisé dans la préparation de l'Ensaimada de Mallorca. On a étudié les propriétés physico-chimiques du saindoux de porc noir de Majorque et son comportement lorsqu'il est soumis à des procédés thermiques. Nous avons évalué l'effet de la source de matières grasses utilisées (lard du bassin rénal* vs *lard sous-cutané) dans la préparation et la qualité de l'Ensaimada, en utilisant le format standard établi par le Conseil de régulation de l'Ensaimada de Mallorca. Les résultats ont montré que les caractéristiques physico-chimiques et le comportement du saindoux de porc noir de Majorque dépendent du type de graisse utilisée dans sa préparation. Le saindoux de porc noir de Majorque a présenté une composition en acides gras mono-insaturés de 45,6% ± 2,6, dans le cas de la graisse du bassin rénal, et de 50,9% ± 3,8, dans le cas de la graisse intermusculaire, principalement en raison de la teneur élevée en acide oléique. En général, le type de gras qui montrait des caractéristiques et un comportement les plus semblables à ce qui est établi par la norme, était celui élaboré avec du lard pelvien rénal.*

Mots-clés. *Porc noir de Majorque – Saindoux –* Ensaimada*.*

I – Introduction

The *Ensaimada de Mallorca* is a typical sweet from the Island of Majorca, made of flour and animal fat, and it is covered by a Protected Geographical Indication. Its name derives from *saïm*, which means pork lard in Catalan, and until the first half of the twentieth century it came exclusively from Majorcan Black Pigs (MBP). Currently, the MBP lard is not used any more in the elaboration of *Ensaimada*. MBP produce high amounts of subcutaneous fat, and most of it is used to produce *Sobrassada*, the main meat product from these animals, the remaining subcutaneous and flare fat has a low market value due to scarce demand for it, thus it is of great interest to enhance the use of subcutaneous fat from MBP to elaborate *Ensaimada*. The first step is to get information regarding the suitability of fat from MBP to elaborate *Ensaimada* determining its physicochemical properties.

The main objective of the present study was to determine the physicochemical properties and thermal behaviour of Majorcan Black Pigs subcutaneous fat to assess its suitability to be used in the elaboration of *Ensaimada de Mallorca*.

II – Materials and methods

Different fat deposits from MBP which could be potentially used to elaborate Ensaimada were sampled at the slaughter plant, including flare fat and intermuscular fat. The quality parameters were studied following the methodologies described below.

Acidity was calculated over the percentage of oleic acid respect to fatty acid profile. Peroxides index express the active oxygen milliequivalents per kg of fat, and it is measured by the potassium iodide oxidation levels. Solid curve was determined by differences in the density when increasing the temperature. The fatty acid profile was determined by gas chromatography and the proportion of each fatty acid group is expressed in percentage. Texture was determined using a TPA analysis using a RA XT 2i Texturometer in fresh samples and the measure was repeated after melting and solidifying each fat sample. Colour was determined by a colorimeter Minolta using the illuminant D65 in fresh fat and melted-solidified samples.

All the measurements were also determined in a reference sample given by the Regulatory Council of *Ensaimada de Mallorca* to compare with the characteristics of flare fat and intermuscular fat from Majorcan Black Pigs.

III – Results

The results obtained showed that the physicochemical characteristics and behaviour of fat from Majorcan Black Pig were dependent on the type of fat tissue used in its preparation (Table 1). The Majorcan Black Pig lard showed a monounsaturated fatty acid composition of 45.6% ± 2.6 in flare fat, and 50.9% ± 3.8 in intermuscular fat, due mainly to high oleic acid content.

IV – Conclusions

The characterisation of the main physicochemical properties and thermal behaviour of the two types of fat showed that flare fat showed more similar traits and behaviour to those presented by the fat sample considered as the reference. It implies that fat from adult Majorcan Black Pigs could be used to elaborate *Ensaimada* maintaining similar properties. Improving the added value of this type of fat would improve economic sustainability of MBP production chain The next step would be to introduce a proportion of intermuscular fat when elaborating *Ensaimada*, to the point of not affecting the technological and sensory traits of the final product achieving the objective of taking as much profit of the fat as raw material.

Acknowledgements

The Authors want to thank Puratos the cooperation received in this study. The research was supported by the Red Rural National grants.

Table 1.- Physicochemical properties and thermal behaviour of flare fat and intermuscular fat from Majorcan Black Pigs and a reference sample from the Regulatory Council of Ensaimada de Mallorca

	Flare fat		Intermuscular fat		Reference	
	Mean	**S.E.**	**Mean**	**S.E.**	**Mean**	**S.E.**
Acidity (% AC OLEIC)	0.42	0.017	0.28	0.021	0.48	0.036
Peroxides	0.59	0.02	0.54	0.035	0.31	0.02
Solid curve (%solids)						
10ºC	48	1.053	36.35	1.586	46.46	2.374
20ºC	32.48	0.989	22.52	1.252	35.24	3.463
30ºC	11.57	0.146	3.5	0.093	15.26	0.704
40ºC	2.64	0.012	0.5	0.031	3.74	0.279
Fatty acids (%)						
SFA	44.03	0.842	39.12	1.533	45.5	2.811
MUFA	45.59	2.593	50.92	3.818	37.9	5.114
PUFA	6.87	0.142	5.95	0.195	13.34	0.409
Texture						
Texture fresh (g)	94.72	0.79	44.31	0.62	83.89	1.995
Texture melted (g)	134.69	0.405	89.8	1.855	127.69	2.168
Color Minolta						
L	87.89	0.017	86.84	0.021	86.59	0.15
a	-1.41	0.011	-2.16	0.029	-1.19	0.031
b	9.28	0.018	8.82	0.03	11.73	0.046
Color Minolta after melting						
L	87.42	0.01	86.64	0.012	84.58	0.087
a	-1.86	0.009	-2.87	0.035	-1.87	0.035
b	7.84	0.009	7.72	0.025	11.9	0.05

Acorn chemical composition depending on shedding date and *Quercus* species

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Abstract. Acorns from *Quercus* species are the main feed resource of free range Alentejano pigs during their fattening period. The animals have access to the acorns as they shed, along the "Montanheira" season. It is well known that chemical composition of acorns vary with species, botanical varieties, harvest year and shedding date. A better knowledge of the chemical composition of acorns along the "Montanheira" may result in a more rational utilization of acorns by pigs and therefore a better management of the "Montanheira". The aim of this study was to evaluate the chemical composition of acorns from Q. *rotundifolia* and *Q. suber*, from October to December. Acorn samples from eight trees were collected from the ground every two weeks (6 collection dates). Acorns were separated into hull and kernel, freeze dried, ground and analyzed for dry matter, ashes, protein, fat, NDF, ADF, starch and tannins. There was an effect of species on the chemical composition, specially for protein content which was about twice as high in the seed of *Q. suber* (83,2 g/kgDM) than in that of *Q. rotundifolia* (39,8 g/kgDM). Date of shedding affected mostly tannin content of acorns, which decreases along maturation process, as it was observed by previous authors.

Keywords. Acorn – Chemical composition – *Quercus rotundifolia* – *Quercus suber* – Shedding date.

Composition chimique des glands en fonction de la date de récolte et l'espèce (*Quercus rotundifolia vs Quercus suber*)

***Résumé.** Le gland de* Quercus rotundifolia *et* Quercus suber *est la principale source de nourriture dans la finition des porcs de l'Alentejo en régime extensif. Les animaux ont accès au gland quand il tombe des arbres au cours de la période de «montanheira». Il est bien connu que la composition chimique des glands varie selon les espèces, les variétés botaniques, l'année de récolte et la date de récolte. Une connaissance plus détaillée de la composition chimique au cours de la «montanheira» peut entraîner une utilisation plus rationnelle des glands et, par conséquent, une meilleure gestion des «montanheira». Le but de cette étude était d'évaluer la composition chimique des glands* (Q. rotundifolia *et* Q. suber*), entre le début octobre et la fin décembre. Des échantillons de glands de huit arbres ont été prélevés sur le sol tous les 15 jours (six dates de récolte), séparés en coquille et graine, lyophilisés, broyés et analysés pour la matière sèche, cendres, protéines, graisse, NDF, ADF, amidon et tanins. Nous avons observé un effet des espèces sur la composition chimique des glands, en particulier pour la teneur en protéines, qui était de 83,2 g / kgMS et de 39,8 g / kgMS pour la graine de* Q. suber *et de* Q. rotundifolia*, respectivement. La date de récolte a affecté la teneur en tanins, qui a diminué au cours du processus de maturation comme précédemment observé par d'autres auteurs.*

***Mots-clés.** Gland – Composition chimique –* Quercus rotundifolia – Quercus suber – *Date de la récolte.*

I – Introduction

Acorns from *Quercus rotundifolia* and *Quercus suber* are, together with pasture, the main feed resources available for the fattening period of Alentejano and Mediterranean pigs in the traditional silvo-pastoral system – "Montanheira" or "Montanera" in Portugal and Spain, respectively. Ripening and shedding of acorns occurs between October and February making the fruits available on the ground to be ingested by the pigs. The acorn chemical composition may vary depending on *Quercus* species and variety, site of production and fruit insect infestation (Gea-Izquierdo *et al*., 2006), among other factors. Shedding date, as a result of fruit maturation, and or climacteric conditions such as hydric stress and strong winds, may also affect chemical composition of acorns and consequently its nutritive value. The chemical composition of acorns has been referred, together with pasture availability, as a determinant factor to the superior quality of meat and meat products from Alentejano and Iberian pigs (López-Bote, 1998; see Tirapicos). "Montanheiras" are carried out, both in *Q. suber*, *Q. rotundifolia* or mixed stands from October to February. Therefore a better knowledge of the chemical composition of acorns associated with these different conditions may contribute to a more rational utilization of "Montanheiras".

The aim of this study was to evaluate the effect of species (*Q. suber* or *Q. rotundifolia*) and time of shedding on the chemical composition of acorns.

II – Material and methods

Eight trees, (four *Q. rotundifolia* and four *Q. suber*) were randomly selected on a mixed oak stand in the Herdade da Mitra (Alentejo, Portugal). A square of about 2 m^2 was defined on the ground, underneath each canopy (crown area) and cleaned with a hand-rake in order to remove all acorns present. Collection of samples (about 500 g) was carried out every fortnight, starting from the day of the first cleaning of the squares. Immediately after each sample collection, the area of the defined squares was cleaned to ensure that each sample contained only acorns fallen during the previous 15 days. Six sample collections were carried out on October the 13th and 27th, November the 10th and 24th and December the 9th and 22nd of 2008. After collection from the ground, the acorns were taken to the laboratory, and a subsample of about 100 acorns was weighed to calculate the average acorn weight. The acorns were than separated into hulls and kernels using a knife and the weight of each portion recorded. Hulls and kernels were frozen at -20 ºC, freeze dried and ground (Ø1mm). Samples of the kernels were analyzed for dry matter (DM) (103º C with forced ventilation for 24 h), ashes (charred followed by combustion in a muffle furnace at 550 ºC), protein by the Dumas method (AOAC, 2005) in a LECO system (FP528), crude fat in a Soxtec system (AOAC, 2005) and neutral detergent fiber (NDF) by the method proposed by Goering and Van Soest (1970) and modified by Van Soest and Robertson (1980). Starch content of kernels was determined using an enzymatic method (kit MEGAZYME K-TSTA 04/2009) as described in AOAC, 2000. Phenolic compounds were extracted from the kernels (100 mg) twice, using an acetone solution at 70% (10 and 5 ml for the first and second extraction respectively). Samples with the acetone solution were sonnicated (20min) at 4 ºC and after centrifuged (3000xg for 10 min at 4 ºC). The supernatants were removed and total phenolic compounds (TPC) were measured using the Folin-Cicaulteau method as described by Makkar *et al*. (1993). Extractable tannins (ET) were calculated indirectly after measuring the non-precipitable phenols (phenols other than tannins, NPP) using polyvinyl polypyrrolidone (PVPP). NPP were left in solution after adding polyvinyl polypyrrolidone (PVPP) to the original extract, which precipitates along with the tannins, and measured as the TPC. ET were calculated as follows: ET=TPC-NPP. The results obtained were submitted to a 2x6 ANOVA (factor 1: oak species; factor 2: shedding period) using the statistics software package SPSS 17.0. Normality of distributions and homogeneity of variances were guaranteed.

III – Results and discussion

Fresh acorn (from Q. *rotundifolia* and Q. *suber*) average weights and percentage of pulp along the six shedding periods are presented in Table 1. Acorn average fresh weight was quite variable, as shown by the sd's presented in Table 1, and ranged from 3.7 to 5.8 g which is within the values reported in the literature (Rodríguez-Estévez *et al*., 2008). Acorns from Q. *rotundifolia* were heavier and larger than those from Q. *suber* ($P<0.05$, Table 2). Although not significant ($P=0.078$) there was a trend to an increase in acorn size along the shedding periods which is in line with an increase ($P<0.05$) in the kernel percentage of the fruits along those periods. The effect of species and shedding period on the physical characteristic of acorns might be relevant for the choice process of the animals for the available acorns, since pigs have been shown to prefer larger acorns with a higher pulp percentage (Rodríguez-Estévez et al., 2008).

Table 1. Physical characteristics of acorns from *Q. rotundifolia* and *Q. suber* along 6 shedding periods (mean±sd)

		Shedding period						
	Acorn	**1**	**2**	**3**	**4**	**5**	**6**	**Total**
Acorn weight (g)	*Q. r.*	3.9±1.5	4.7±1.0	5.5±1.7	5.8±0.8	5.7±0.5	5.3±0.4	5.1
	Q. s.	3.9±0.7	3.7±0.6	4.1±0.2	4.9±1.1	4.8±1.3	4.1±1.4	4.3
	Total	3.9	4.2	4.8	5.3	5.3	4.7	4.7
Acorn pulp (%)	*Q. r*	64.7±10.9	69.9±12.2	75.1±9.0	79.4±4.0	77.9±1.4	78.3±1.3	74.2
	Q. s	65.1±8.5	65.0±10.2	74.9±5.2	79.7±1.6	80.3±1.8	81.1±3.6	74.3
	Total	64.9^{a}	67.5^{a}	75.0^{ab}	79.6^{b}	79.1^{b}	79.7^{b}	74.3

[a-c] Means within the same row with no common superscript are significantly different (P<0.05) according to Tukey's HSD test. *Q.r.: Quercus rotundifolia; Q.s.: Quercus suber.*

Table 2. Statistical parameters for the analysis of the effect of species and shedding period on the physical characteristics of acorns

	Effects								
	Species			**Shedding period**			**SpxShed.p**		
	Sig[1]	**SEM[2]**	**n**	**Sig**	**SEM**	**n**	**Sig**	**SEM**	**n**
Acorn weight (g)	0.01	0.22	24	0.078	0.38	8	0.836	0.55	48
Acorn pulp (%)	0.941	1.43	24	0.00	2.48	8	0.907	3.51	48

[1]Significance; [2] Standard error of the mean.

Chemical composition of acorn kernels from *Q. rotundifolia* and *Q. suber* along the six shedding periods is shown in Table 3. Data obtained is within the range of values previously presented in the literature (Gea-Izquierdo *et al*., 2006, Tirapicos Nunes, 2007).

Variable nutrient contents of acorns, even within the same species, have been reported by Gea-Izquierdo *et al*. (2006) and are also present in our data. Acorn species had a significant effect on its kernel chemical composition (Table 4). Overall, kernel from *Q. rotundifolia* had a lower content in ashes and a higher content in fat, NDF and starch than that from *Q. suber* (Table 3). Protein was the nutrient that showed the largest difference between species. Although still low in protein, Q. *suber* kernels may supply twice as much protein when compared to *Q. rotundifolia* (83.7 *vs* 39.4 g/kg DM), which might be important in the absence or lack of pasture under the "Montanheira" system. Dry matter content of kernels from *Q. suber* was lower (P<0.05), than that from *Q. rotundifolia*. This was probably due to the earlier stage of maturation evidenced by the presence of a larger proportion of green acorns for *Q. suber* than for *Q. rotundifolia* in the first two or three shedding periods. Shedding period also affected (p<0.05) the DM content of acorns which is in line with the observed increase in its major components, fat and starch. Dry matter content of acorns is also highly dependent on the environment humidity (Vázquez *et al*., 2001), however this parameter was not registered in the present work.

Shedding period did not affect ash and protein content of acorn kernels (Table 4). This is not in agreement with Carbonero *et al*., (2006) that observed an increase in protein content along the Montanheira period. Starch is the major constituent of acorn kernel and its concentration increased (P<0.05) as the "Montanheira" progressed. A significant effect (P<0.05) of shedding period was also observed on kernel fat and NDF content. Fat content was higher than that reported by other authors (Almeida *et al*., 1992; Carbonero *et al.*, 2006) but also increased during the "Montanheira" period (on average from 59.5 to 99.3 g/kg DM).

Table 3. Chemical composition of acorn kernel from Q. *rotundifolia* and Q. *suber* along 6 shedding periods

		Shedding period						
		1	**2**	**3**	**4**	**5**	**6**	**Total**
DM g/kg	*1*	472,8±98,8	551,2±99,1	599,8±33,2	627,5±17,1	600,9±20,6	594,8±15,5	574,5
	2	358,0±76,0	426,2±76,0	493,3±43,3	554,6±20,0	549,8±6,7	544,4±15,7	487,7
	T	415,4a	488,7ab	546,6bc	591,1c	575,4c	569,6bc	531,1
Ashes g/kgDM	*1*	21,4±1,5	20,7±2,1	21,1±1,6	21,8±1,7	19,4±3,1	21,9±0,9	21,1
	2	28,8±3,6	29,8±3,3	29,2±2,3	29,0±5,1	25,1±1,8	25,8±1,9	27,9
	T	25,1	25,6	25,1	25,4	22,3	23,9	24,5
Protein g/kgDM	*1*	37,7±5,0	37,7±6,0	35,6±6,7	42,4±6,3	43,4±6,3	39,6±6,0	39,4
	2	93,8±7,9	87,0±7,5	86,3±5,0	83,4±6,6	75,7±8,2	76,0±10,5	83,7
	T	65,8	62,3	60,9	62,9	59,6	57,8	61,6
Fat g/kgDM	*1*	83,8±31,9	96,8±31,8	135,1±16,8	122,0±6,0	114,6±12,2	116,3±18,0	111,4
	2	35,1±20,8	48,2±20,8	68,5±13,6	79,0±12,1	82,0±20,8	82,2±15,7	65,8
	T	59,5a	72,5ab	101,8c	100,5bc	98,3bc	99,3bc	88,6
NDF g/kgDM	*1*	45,0±4,1	55,0±12,5	73,9±3,1	56,9±19,1	50,1±9,5	50,6±4,9	55,3
	2	45,0±4,0	49,1±3,7	58,9±3,8	50,7±18,8	42,4±1,0	39,4±2,7	47,9
	T	45,0a	52,1a	66,4b	53,8ab	46,3a	45,0a	51,4
Starch g/kgDM	*1*	512,5±19,9	479,8±43,3	522,1±28,3	599,5±10,8	551,2±17,9	535,1±46,8	536,1
	2	220,3±19,4	357,0±69,0	410,9±25,9	536,0±28,9	508,7±44,3	510,4±44,3	430,8
	T	386,5a	418,4ab	466,5b	567,8c	530,0c	531,7c	483,5

[a,b] Means within the same row with no common superscript are significantly different (P<0,05) according to Tukey's test. 1*: Quercus rotundifolia;* 2*: Quercus suber*.

Concentrations of phenolic compounds measured as g of tannic acid equivalent per kg sample, are shown in Table 5. Extractable tannins represented, on average, 97% (ranging from 93-98%) of the total phenolic compounds measured, suggesting that only a very small proportion of those were non-precipitable phenols. Kernels from *Q. suber* had a higher concentration of phenolic compounds then those from *Q. rotundifolia*, furthermore, phenolic compounds concentration of both species decreased (P<0.05) with the progression of "Montanheira". This has also been reported by Almeida *et al*. (1992).

Table 4. Statistical parameters for the analysis of the effects of species and shedding period on the chemical composition of acorns

	Effects								
	Species			Shedding period			SpxShed.p		
	Sig[1]	SEM[2]	n	Sig	SEM	n	Sig	SEM	n
DM	0,00	11,14	24	0,00	19,30	8	0,61	27,29	48
Ashes	0,00	0,54	24	0,16	0,94	8	0,44	1,33	48
Protein	0,00	1,43	24	0,31	2,47	8	0,01	3,49	48
Fat	0,00	3,95	24	0,00	6,84	8	0,82	9,67	48
NDF	0,01	1,92	24	0,00	3,33	8	0,71	4,71	48
Starch	0,00	8,50	24	0,00	14,72	8	0,00	20,82	48

[1]Significance; [2] Standard error of the mean.

Table 5. Concentration of phenolic compounds (g standard equivalen/kg acorn kernel) on acorn kernel from Q. *rotundifolia* and Q. suber along 6 shedding periods

		Shedding period						
		1	2	3	4	5	6	Total
TPC[1]	*Q. rot*	57,5±10,2	42,8±10,1	37,7±3,4	28,7±2,6	33,3±4,7	38,1±5,1	39,7
	Q. suber	129,5±18,6	85,1±20,5	67,9±8,5	47,6±7,7	58,0±14,8	63,0±16,7	75,2
	Total	93,5a	63,9b	52,8bc	38,2c	45,7c	50,6bc	57,4
ET[2]	*Q. rot*	55,0±10,3	41,0±10,3	36,0±3,5	27,6±2,7	32,0±4,6	36,6±5,2	38,1
	Q. suber	126,3±18,9	83,2±18,3	66,5±8,3	46,4±7,6	56,5±14,6	61,5±16,4	73,4
	Total	90,7a	62,1b	51,3bc	37,0c	44,3c	49,0bc	55,7

[a-c] Means within the same row with no common superscript are significantly different (P<0,05) according to Tukey's HSD test; [1] Total phenolic compounds; [2] Extractable tannins.

Table 6.Statistical parameters for the analysis of the effects of species and shedding period on the chemical composition of acorns

	Effects								
	Species			Shedding period			SpxShed.p		
	Sig[1]	SEM[2]	n	Sig	SEM	n	Sig	SEM	n
TPC[3]	0,00	2,34	24	0,00	4,06	8	0,00	5,74	48
ET[4]	0,00	2,35	24	0,00	4,06	8	0,00	5,75	48

[1]Significance; [2] Standard error of the mean; [3] Total phenolic compounds; [4] Extractable tannins.

IV – Conclusions

Chemical composition of acorn kernels is quite variable and this can be observed even within the same species. Acorn species affect their chemical composition in that those from *Q. rotundifolia* have a lower content in ashes, protein and tannins and a higher content in fat, NDF and starch than that from *Q suber*. These results show the nutritional advantage of *Q. rotundifolia* acorns, especially in what concerns to its energetic value due to the higher concentrations in starch and fat. These differences in chemical composition together with the lower concentration on tannins may justify the commonly observed preference of pigs for *Q. rotundifolia* acorns. Shedding period also affected the chemical composition of acorn kernels. In fact, an improvement of the nutritive value was observed, with fat, NDF and starch content increasing and tannin content decreasing as the "Montanheira" progressed.

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Session 4
Health

Iberian pig raising: Animal health and food safety

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Abstract. Food safety – the ensuring that foods are safe, healthy and of high quality – is an essential feature of food manufacture, and animal health one of the most important features of stock-raising. The 2007-2013 strategy of the European Union promotes animal health via the prevention and/or reduction of the incidence of certain animal diseases with the aim of guaranteeing public health and food safety. The control of animal disease is therefore vital, especially of infections/infestations that are transmissible (either directly or indirectly) to humans (zoonoses). As part of the projects, "Saniberico 08/222" (Corporación Tecnológica de Andalucía, Consejería de Innovación, Ciencia y Empresa, Junta de Andalucía) and "IDI-20090414" (Spanish Ministerio de Ciencia e Innovación), we undertook the study "Assessment and improvement of Iberian pig health and food product safety" to determine the design needs for an animal health and food safety program based on HACCP, with special attention paid to Salmonella, Brucella, Toxoplasma and Trichinella. In this communication the results of this pioneering research project in our country are presented and shared with researchers from the international scientific community, with the final aim of promoting and spread information about food safety and quality excellence of products derived from the Iberian pig.

Keywords. Iberian pigs – Zoonoses – Animal health – Food safety.

Santé animale et sécurité alimentaire chez le porc Ibérique

Resumé. *La sécurité alimentaire – la garantie que les aliments sont sûrs, sains et de haute qualité – est une caractéristique essentielle de la fabrication des aliments, et une des caractéristiques les plus importantes de l'élevage. La stratégie 2007-2013 de l'Union européenne favorise la santé animale par le biais de la prévention et / ou la réduction de l'incidence de certaines maladies animales dans le but de garantir la santé publique et la sécurité alimentaire. Le contrôle des maladies animales est donc essentiel, en particulier des infections / infestations qui sont transmissibles (soit directement ou indirectement) à l'homme (zoonoses). Dans le cadre des projets "Saniberico 08/222" (Corporación Tecnológica de Andalucía, Consejería de Innovación, Ciencia y Empresa, Junta de Andalucía) et "IDI-20090414" (Ministère de la Science et de l'Innovation de l'Espagne), nous avons entrepris une étude pour "l'évaluation et l'amélioration de la santé du porc Ibérique et la sécurité des produits alimentaires" pour déterminer les besoins de conception d'un programme de santé animale et de salubrité des aliments basé sur le HACCP, avec une attention particulière à* Salmonella, Brucella, Toxoplasma *et* Trichinella*. Dans cette communication, les résultats de ce projet de recherche, pionnier dans notre pays, sont présentés et partagés avec des chercheurs de la communauté scientifique internationale, dans l'objectif final de promouvoir et de diffuser des informations sur la sécurité alimentaire et l'excellence de la qualité des produits dérivés du porc Ibérique.*

Mots-clés. *Porc Ibérique – Zoonoses – Santé animale – Sécurité alimentaire.*

Food safety – the ensuring that foods are safe, healthy and of high quality – is an essential feature of food manufacture, and animal health one of the most important features of stock-raising. The 2007-2013 strategy of the European Union promotes animal health via the prevention and/or reduction of the incidence of certain animal diseases with the aim of guaranteeing public health and food safety. The control of animal disease is therefore vital, especially of infections/infestations that are transmissible (either directly or indirectly) to humans (zoonoses).

Attempts to establish surveillance and control measures for zoonotic diseases in Iberian pigs could improve the safety of their products. For such systems to be put in place, information is required on the current disease status, as well as the risk factors for disease, the critical steps in the food manufacturing chain, and the interactions of these variables. Practical, effective control strategies adapted to the peculiarities of Iberian pig production are also required.

The raising of Iberian pigs in the dehesa ecosystem (grassland plus Mediterranean oak), where acorns are a major part of the diet, exposes these animals to environmental conditions that favour the dissemination of pathogens, the control of which is difficult. In these extensive or semi-extensive systems the animals are in close contact with one another, with the environment, and with other animals, particularly rodents, peridomestic animals, and wild animals such as foxes, wild boar and birds. Together, these can play important roles in the epidemiology of *Salmonella* spp., *Campylobacter* spp., *Brucella* spp., Aujeszky's disease, classical swine fever (CSF), African swine fever (ASF), tuberculosis, etc. The recent outbreaks of ASF in Europe posed a serious threat to Iberian pig raising; if the disease had not been controlled it could have entered the dehesa system and been maintained among wild boar. Disease among pig stocks would have led to the sector suffering marketing restrictions.

Iberian pigs farms have different structures, ranging from small or family interests (sometimes with very deficient infrastructures) to exploitations with high zootechnic performance and very good infrastructures. Adequate perimeter fencing and 'health firebreaks' play a vital role in preventing the entry and dissemination of pathogens, yet only 35% of farms are thus protected.

The structural diversity of Iberian farms, and the many small exploitations that exist, is reflected in the diversity of animal health programs in operation. Some studies report that over 40% of farmers have no animal health program in place at all, vaccinating none of their stock against any disease. In the remaining farms, vaccinations against at least two diseases are provided, usually against erysipelas and pasteurellosis. The latter two diseases plus parvovirosis, clostridiosis, colibacilosis, PRRS, Aujeszky's disease (officially included in the Spanish National Plan for the Eradication of Animal Diseases) and infection by *Mycoplasma* spp. form the range against which animals are usually protected.

Health status is one of the most important variable costs to affect the performance of exploitations; poor health prevents animals reaching their maximum productive potential since it influences their feed intake, daily weight gains and feed conversion indices. Clinical disease incurs direct costs associated with medicines and vaccines, etc., while subclinical infections generate indirect costs via the reduction in main daily weight gain, greater protein and energy needs, and poorer metabolism. In some cases it is also associated with losses occasioned by rejection at slaughter.

The wide infrastructural and health status diversity of Iberian pig farms renders it very difficult to determine the incidence of different diseases in this industry. However, some epidemiological data are available, showing that Aujeszky's disease is present on 15% of exploitations, and that at least 20% of Iberian pigs suffer erysipelas (the causal agent *Erysipelothrix rhusiopathiae* remaining latent in these animals' tonsils). The incidence of other infections/diseases can be estimated via the diagnosis of clinical processes in animals, although it should be remembered these data are available only when clinical episodes are recognised.

Clinical processes should be classified according to the clinical problems they cause, e.g., miscarriages, respiratory problems and intestinal problems. *P. multocida*, either alone or in conjunction with other pathogens, is the main agent involved in clinical respiratory problems. *E. coli* and *Cl. perfringens* type C are common causes of intestinal disease in piglets, while in fattening pig's parasites and porcine dysentery are the major problems. Underlying infection with porcine circovirus type 2 (PCV-2), an immunodepressing pathogen, can cause the incidence of respiratory and/or intestinal infections to increase. Reproductive problems in Iberian pigs are mainly caused by viruses, e.g., pavovirosis. *Chlamydiophila* spp. and leptospirosis are also responsible for miscarriages.

The guts of animals are the main reservoir of *Salmonella*, although usually there are many carriers but few sufferers of disease. *Salmonella* is able to multiply over a wide range of temperatures and to survive long periods in water and organic substrates such as manure. This persistence in the environment provides a source of contamination for birds, rodents and arthropods, etc., which then contribute to the indirect maintenance of the bacterium in farm animals and its circulation among them.

Within the framework of the project "Saniberico 08/222" (Corporación Tecnológica de Andalucía, Consejería de Innovación, Ciencia y Empresa, Junta de Andalucía), our group studied the prevalence of *Salmonella* in Iberian pig farms in southern Spain in 2008/09, making checks at slaughterhouses. The pigs involved came from 81 farms, 67 of which were free range (as classified by Spanish Royal Decree 1469/2007). A wide diversity of seroptypes and phagotypes was detected. The prevalence data showed low infection indices (5.3% of animals positive), although 33% of the farms carried infected animals. The most common serotypes were Anatum and Typhimurium. A number of uncommon serotypes were also isolated, such as Hessarek and Mikawasima, associated with wild animal reservoirs such as foxes, rodents and wild birds (turtle doves, doves, corvids). The isolated strains were subjected to testing against 16 antimicrobial agents and showed only low-level resistance, except against streptomycin (46%), tetracycline (30%), sulphonamide (25%) and ampicillin (23%). The number of multi-resistant strains (i.e., resistant to ≥4 agents) was moderate (36%). The prevalence figures quoted above are similar to those reported by our group for intensively-raised white pigs, although with significant differences in terms of serotypes, phagotypes and microbial resistance.

During 2009-10, we examined 709 serum samples from 10 Iberian pig farms with different systems (acorn feed, mixed feed, and commercial feed). These samples were obtained at slaughter and kept at –20ºC until their analysis using different commercial ELISA kits to detect *Brucella*, *Salmonella*, *Toxoplasma* and *Trichinella*. *Salmonella* infection and *Toxoplasma* infestation were found to be amply distributed among the studied farms (73% and 54%, respectively); no such findings were made for the other pathogens screened (e.g., *Brucella* 3.8% and *Trichinella* 0%).

Campylobacter spp. also causes zoonoses. The main reservoir for this pathogen is the digestive tract of wild and domestic mammals and birds, which, via their faeces, contaminate the ground and water sources. *Campylobacter* shows the ability to persist in water.

Other infections of Iberian pigs include those caused by mycobacteria, *Rhodococcus equi*, *Streptococcus* spp., *Staphylococcus* spp. and *Arcanobacterium* spp. All of these can cause chronic caseous or purulent lymphadenitis (granulomas/pyogranulomas), affecting the lymph glands and viscera. Signs of infection are also visible in carcasses after slaughter, leading to their rejection and economic losses. Although the prevalence of these processes can vary widely from one farm to another, it is estimated that up to 50% of all rejections are owed to this type of lesion. Infection by the above agents, which is considered very infrequent in commercial pigs, is facilitated in free range Iberian pigs by the conditions on extensive and semi-extensive farms, i.e., through contact with wild animals acting as reservoirs of these pathogens.

Many of the above diseases are zoonotic in nature, but while they may be subclinical in animals they can be of great public health importance. It is therefore important to understand the current disease status of each and to establish risk factors and identify critical points in the food manufacturing chain (farms, slaughterhouses and the meat industry).

As a primary health requirement, Iberian pig farms should aspire to be free of notifiable diseases such as swine flu, porcine dysentery, PRRS, and those caused by *Salmonella, Actinobacillus pleuropneumoniae, Mycoplasma hyopneumoniae, Leptospirosis,* PCV2, *Bordetella bronchiseptica, Pasteurella multocida, Chlamydiophila psittaci* and *Ascaris suum*, as well as porcine proliferative enteritis.

Finally, all forms of Iberian pig farms (acorn feed, mixed feed and commercial feed) should undertake the following:

In the area of animal health:

1.- The detection of common diseases and production deficits.

2.- The implantation of preventive medicine programs that guarantee animal health and profitability.

3.- The implantation of biosafety measures, including protocols to verify these measures, appropriate for the different types of farm, aimed at optimising mid-long term animal health.

4.- The selection of health-certified replacement animals.

In the area of public health:

1.- To determine the presence of zoonoses of particular interest to public health (salmonellosis, listeriosis, tuberculosis, brucellosis, campylobacteriosis, trichinellosis, toxoplasmosis, echinococcosis, cysticercosis), as well as pyrogranulomatous processes caused by *Archanobacterium* and *Rhodococcus*. The diffusion of these problems in herds should be checked.

2.- To design a pre-slaughter program for the improvement of animal health and food safety based on descriptive epidemiological studies of potential zoonotic agents, hazard analysis and the identification of critical control points (HACCP), and the establishment good hygienic practice codes throughout the food production chain.

As part of the projects ‘Saniberico 08/222’ (Corporación Tecnológica de Andalucía, Consejería de Innovación, Ciencia y Empresa, Junta de Andalucía) and ‘IDI-20090414’ (Spanish Ministerio de Ciencia e Innovación), we undertook the study “Assessment and improvement of Iberian pig health and food product safety” to determine the design needs for an animal health and food safety program based on HACCP, with special attention paid to Salmonella, Campylobacter, Listeria, Brucella, Toxoplasma and Trichinella.

Serological study of potential zoonotic pathogens in Iberian pigs

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Abstract. Zoonotic diseases can course as inapparent diseases in porcine livestock, however, they present a significant impact in the field of public health. These diseases include *Brucella* and *Salmonella* infections and infestations with *Toxoplasma* and *Trichinella*, all considered as high zoonotic risk agents by the EFSA (2010). For this reason, we performed a serological screening which allows us to know indirectly the presence and distribution of these pathogens in Iberian pigs. A total of 709 sera from 79 farms in the Iberian pig, reared in different systems were collected at slaughterhouses and kept at -20ºC until analysis. Sera were analysed with different antigens by commercial ELISA kits: *Brucella* (Ingenasa), *Salmonella* (Svanova), *Toxoplasma* (IDVet) and *Trichinella* (Prionics). The results obtained in our study allow us to conclude that *Salmonella* infection and infestation by *Toxoplasma* are widely distributed in the farms tested (73.42% and 54.43% respectively), unlike other pathogens checked, as *Brucella* and *Trichinella*, which showed low serological prevalence (3.8% and seronegative, respectively).

Keywords. Iberian pigs – ELISA – Zoonotic – Seroprevalence.

Étude sérologique de pathogènes potentiellement zoonosiques chez le porc Ibérique

Résumé. *Les maladies à caractère zoonosique peuvent suivre un cours de façon inaperçue chez le bétail porcin, mais ont cependant une répercussion très importante sur le domaine de la santé publique. Il faut souligner –parmi ces processus– les infections par* Brucella *et* Salmonella *et les infestations par* Toxoplasma *et* Trichinella*. Toutes ces maladies sont considérées à haut risque zoonosique selon l'EFSA (2010). Pour cette raison, nous avons réalisé un screening sérologique qui nous permet de connaître d'une façon indirecte la présence et la diffusion de ces agents chez le porc Ibérique. Un total de 709 sérums, originaires de 79 fermes de porcs Ibériques élevés selon différents systèmes, ont été obtenus dans l'abattoir et conservés à -20ºC, jusqu'au moment de leur analyse. Les sérums ont été confrontés à différents antigènes au moyen de kits commerciaux ELISA :* Brucella *(Ingenasa),* Salmonella *(Svanova),* Toxoplasma *(IDVet) et* Trichinella *(Prionics). Les résultats obtenus dans notre étude permettent de conclure que l'infection par* Salmonella *et l'infestation par* Toxoplasma *sont toutes deux amplement répandues dans les fermes analysées (73,42% et 54,43%, respectivement) ; au contraire,* Brucella *et* Trichinella *ont peu de prévalence sérologique (3,8% et séronégatif, respectivement).*

Mots-clés. Porc Ibérique – ELISA – Zoonoses – Séroprévalence.

I – Introduction

Zoonoses are diseases which are transmissible from animals to humans and viceversa. The modernization of the agrifood industry and the growing demand of knowledge from consumers about what do they eat has increased the necessity of control measures to obtain safe and healthy products from animals. Data from the World Health Organization (WHO) confirmed that in the last 10 years about the 75% of human diseases has been related to the presence of pathogens in products from animal origin (EFSA, 2010).

Pork is the most consumed meat per capita in Europe, followed by poultry and cattle meats (Eurostat, 2008). Spain occupies the second place in both pig and pork production and is considered as the highest pork consumer between the 27 member states of Europe (Marquer, 2010). Thus, all those aspects concerning pig health and pork safety are nowadays of significant interest first in the world market, but even with higher emphasis in Spain.

Pig livestock represents a potential risk for zoonoses due to both bacterial and parasitic pathogens. These agents may infect humans by different routes, being the most significant one the oral route or consumption of contaminated meat. Zoonotic diseases usually course as inapparent diseases in porcine livestock, displaying a significant impact in the field of public health. These diseases include *Brucella* and *Salmonella* infections and infestations with *Toxoplasma* and *Trichinella*, all considered as high zoonotic risk agents by the EFSA (2010).

The status "pork free of zoonotic agents" may favor international trades giving a differential attribute and positioning in exigent markets such as those from United States and Japan. The each time more frequent and demanded exports of Iberian pig products to foreign countries are equally affected by this situation. For this reason, a serological screening was carried out to determine indirectly, by means the detection of specific antibodies, the prevalence of these pathogens in Iberian pigs reared in different systems.

II – Materials and methods

Seventy-nine Iberian pig herds from southern Spain were randomly selected and sampled during 2008 and 2009. Sample size was assessed by the software Win Episcope version 2.0 on the basis of the number of samples required for a previous unknown prevalence (95% confidence level and 8% accepted error were assumed and confidence intervals of the prevalence were calculated).

Iberian pig herds consisted of both free-range and intensive systems, being classified within three different categories depending on the rearing system and nutritional habits (Table 1):

- Montanera (acorn-fed, AF): the animals are reared in free-range systems and they are fed only by natural resources (i.e. acorn, pasture). This period goes from October to March (next year), beginning the animals with a weight of 92-115 kg, and with a minimum average weight gain of 46 kg and a minimum period of occupation of 60 days.

- Recebo (mixed-fed, MF): the animals are reared in free-range systems but they are fed by both natural resources and commercial feed. This type of nutrition is used when there are not enough natural resources to guarantee that the animals reach the optimal weight to be slaughtered being fed only by natural resources.

- Cebo (commercial feed, CF): the animals may be reared in outdoor facilities or in intensive systems and they are fed only by commercial feed.

Table 1. Distribution of the herds and animals analysed in the different systems

	AF	MF	CF	Total
Herds	63	7	9	79
Animals	564	70	75	709

AF: acorn-fed; MF: mixed-fed; CF: commercially feed

Five to ten pigs per herd were randomly sampled, and blood samples from each were collected at the slaughterhouse. Samples were collected into evacuated tubes, allowed to clot at room temperature and centrifuged, and then the serum was harvested and frozen at -80 ºC until testing

Sera samples were analyzed for the detection of specific antibodies against *Salmonella* spp., *Brucella* spp., *Trichinella* spp. and *Toxoplasma gondii* by means of commercial enzyme-linked immunosorbent assay (ELISA) kits, following manufacturer's instructions (SALMOTYPE® Pig Screen+E, Labor Diagnostik Leipzig; Ingezim Brucella Compac 2.0, Ingenasa; PrioCHECK® Trichinella Ab, Prionics; ID Screen® Toxoplasmosis Indirect, IDVet Innovative Diagnostics). The cut-off value used for discriminating between positive and negative serum samples was 40% for *Salmonella* spp. and *Brucella* spp. antibodies, 15% for *Trichinella* spp. antibodies and 50% for *Toxoplasma gondii* antibodies.

Magnetic stirrer method for pooled sample digestion was also performed on all the animals sampled following the regulation EC-2075/2005. Briefly, one hundred grams of samples at a time (one gram from diaphragm pillar per animal) were chopped in the blender, added to the digestion fluid (1% hydrochloric acid solution + 1% pepsin in tap water) and stirred on a magnetic stirrer for 30-60 minutes at 44-46 ºC. The digest was allowed to settle for 15–20 minutes and the upper two-thirds of the fluid was decanted. The remaining fluid and deposit were poured through a 355 µm mesh screen into a conical settling glass and allowed to settle for a further 15-20 minutes. The maximum possible supernatant fluid was aspirated without disturbing the sediment, and the latter was washed with warm (37°C) tap water and allowed to settle for another 15-20 minutes. The washed sediment was transferred to a 50-ml tube, allowed to settle, and aspirated down to a final volume of 10 ml. All 10 ml were poured into a gridded Petri dish and examined for *Trichinella* larvae with a dissecting microscope (x15-40 magnification).

Since the ELISA test and the artificial digestion yielded contradictory results to detect *Trichinella* spp., an alternative second ELISA test (ID Screen® Trichinella Indirect, IDVet Innovative Diagnostics; cut-off > 50%) was used in order to confirm the results.

III – Results

Specific antibodies against *Salmonella* spp. and *Toxoplasma gondii* were widely distributed, whereas specific antibodies against *Brucella* spp. and *Trichinella* spp. were scarcely detected. No significant differences were observed between the different rearing systems examined, however, the number of mix-fed and fed herds was not enough representative to obtain an accurate estimation.

Antibodies against *Brucella* spp. were found only in 3 out of 63 AF herds (4.76% of herd prevalence), being detected in 6 out of 564 animals (1.06% of individual prevalence). *Salmonella* spp. was more spread, being detected in all the three rearing systems with a herd and individual prevalence close to or higher than 70% and 20%, respectively (Tables 2 and 3).

Toxoplasma gondii-specific antibodies were also widely detected in all the three systems (AF, MF, and CF herds), showing a total herd prevalence of 54.43% and a total individual prevalence of 27.12%. Only three animals (1 animal per herd) showed antibodies against *Trichinella* spp. (Tables 2 and 3). However, all the sampled animals displayed negative results for routine artificial digestion. The alternative second ELISA test used to confirm those contradictory results yielded also negative results.

IV – Conclusions

The results presented in this research showed a high herd and individual prevalence for both *Salmonella* spp. (73.42% and 20.87%, respectively) and *Toxoplasma gondii* (54.43% and 27.12%, respectively) antibodies, without significant differences between the different rearing systems examined. Contrary, prevalence levels for specific antibodies against *Brucella* spp. and *Trichinella* spp. were low.

Table 2. Number of positive herds and prevalence data against each zoonotic pathogen

	Number of positive animals (prevalence)					
				Trichinella		
	Brucella	***Salmonella***	***Toxoplasma***	**ELISA 1**	**ELISA 2**	**Artificial digestion**
AF (n=63)	3 (4.76)	44 (69.84)	37 (58.73)	1 (1.59)	0 (0.00)	0 (0.00)
MF (n=7)	0 (0.00)	7 (100)	2 (28.57)	0 (0.00)	0 (0.00)	0 (0.00)
CF (n=9)	0 (0.00)	7 (77.78)	4 (44.44)	2 (22.22)	0 (0.00)	0 (0.00)
Total Herds (n=79)	3 (3.80)	58 (73.42)	43 (54.43)	3 (3.80)	0 (0.00)	0 (0.00)

AF: acorn-fed; MF: mixed-fed; CF: commercially feed

Table 3. Number of positive animals and prevalence data against each zoonotic pathogen

	Number of positive animals (prevalence)					
				Trichinella		
	Brucella	***Salmonella***	***Toxoplasma***	**ELISA 1**	**ELISA 2**	**Artificial digestion**
AF (n=564)	6 (1.06)	112 (19.86)	171 (30.32)	1 (0.18)	0 (0.00)	0 (0.00)
MF (n=70)	0 (0.00)	20 (28.57)	3 (4.29)	0 (0.00)	0 (0.00)	0 (0.00)
CF (n=75)	0 (0.00)	16 (21.33)	18 (24.00)	2 (2.67)	0 (0.00)	0 (0.00)
Total Animals (n=709)	6 (0.85)	148 (20.87)	192 (27.12)	3 (0.42)	0 (0.00)	0 (0.00)

AF: acorn-fed; MF: mixed-fed; CF: commercially feed

A high seroprevalence of *Salmonella* and *Toxoplasma*, together with minimal prevalence of *Trichinella*, has been previously reported at conventional swine production systems (Gebreyes *et al*., 2008), being observed also a higher prevalence of these pathogens in porcine reared in outdoor-herds compared to those reared in indoor-herds. In our study no significant differences were observed between the different rearing systems, however, the low number of herds sampled for MF and CF herds did not allow us to validate our results. Therefore, further studies should be conducted to carry out a more accurate approach concerning the seroprevalence in different rearing systems.

Mejía *et al.* (2006) reported a 77% of *Salmonella* herd seroprevalence in finishing pig units in Northern Spain. In addition, in a study carried out in our same geographical area a slightly higher individual (27.3%; CI_{95}: 26.1-28.4) and herd seroprevalence (80.0%; CI_{95}:71.5-86.9) of *Salmonella* was observed in pigs reared in intensive systems (Pérez-Barrios *et al*., 2010). These results are in agreement with the results presented in the present study.

Seroprevalence of *Toxoplasma gondii* has been documented worldwide, showing a frequent contact with parasite within the porcine livestock (García-Bocanegra *et al*., 2010; Hill *et al*., 2010; Veronesi *et al*., 2010). In Spain, a previous study has reported a widespread, but variable, seroprevalence of *Toxoplasma gondii* ranging from 4.4% to 27.3% in domestic pigs (García-Bocanegra *et al*., 2010). All-in all-out, cleaning measures, age, control of rodents and cats and carcass disposal methods have been related to differences in *Toxoplasma gondii* seroprevalence (García-Bocanegra *et al*., 2010; Hill *et al*., 2010; Veronesi *et al*., 2010).

Interestingly, in our study ELISA test for *Trichinella* spp. showed positive results for only three animals, however, all the animals displayed negative results by the reference method of detection (magnetic stirrer method for pooled sample digestion) (EC-2075/2005). Moreover, those doubtful results yielded also negative results by the diriment alternative ELISA test.

ELISA represent a useful a rapid method to detect the presence of specific antibodies on serum, plasma or meat juice collected before or after slaughter. *Trichinella* infestation levels as low as one larva/100 g of tissue have been detected by ELISA in pigs (Gamble *et al*., 2004). This high level of sensitivity point to ELISA as a useful method for detecting ongoing transmission of *Trichinella* spp. infection at the farm or for surveillance programmes. Although ELISA kits usually have a high sensitivity, some false-negative results may be observed due to infected animals do not develop an antibody response until 3-5 weeks post infection (Gamble, 1996). In addition, ELISA may yield a low rate of false-positive results due to the specificity of ELISA for *Trichinella* infection is variable according to the type and quality of the antigen employed in each test (OIE, 2009). This results are in agreement with the results obtained in our study and may explained the differences observed between the two ELISA kits used to detect *Trichinella* spp.-specific antibodies. For this reason, serological tests are only recommended for surveillance studies, whereas direct methods should be used to individual carcass inspection (OIE, 2009).

Interestingly, all the pathogens examined in this study has been previously found in wild boars showing similar seroprevalence trends (Gauss *et al*., 2005; Montagnaro *et al*., 2010). Therefore, measures to control the contact between domestic and wild species are especially encouraged in the plans oriented to diminish the prevalence of these pathogens.

Our results showed a wide dissemination of *Salmonella* spp. and *Toxoplasma gondii* in finishing Iberian pigs, whereas *Brucella* spp. and *Trichinella* spp. were rarely or not found. Due to the potential zoonotic risk of these pathogens, which represent a risk both at farm level and at pork level, and the present consumer's concern about food safety control measures should be adopted in order to diminish their prevalence from farm-to-fork.

Acknowledgements

The authors wish to acknowledge the financial support from COVAP, Andalusian Government (ISC, Counseling) (SANIBERICO 08/222), and Centre for the Development of Industrial Technology, CDTI (IDI-20090414).

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Litter size and health management as limiting factors of "*in situ*" conservation of Turopolje pig

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Abstract. Since 1996, the Turopolje pig is recognised as a Croatian autochthonous extensive pig breed and was include in program of "*in situ*" preservation. The purpose of this paper was to analyze the changes in breeding population of Turopolje pig in relation to reproductive traits and brucellosis outbreaks. 529 liters from 388 sows were analysed in the period from 2000 to 2009. In average 1.2 liters/sow/year were observed with 4.21 and 3.90 piglets born alive and weaned per litter, respectively. Mortality rate during the weaning period was close to 20%. In the period from 1996 to 2009 the size of breeding population was increased more than ten times in the number of sows and five times in the number of boars. In 2008, the numbers of sows and boars decreased about 21% in relation to 2007, primarily due to an outbreak of brucellosis. In average 44.2% and 30.1% of tested pigs during 2008 and 2009 were serologically positive to *Brucella suis* infection and were excluded from population. These results suggest requirements to increase the number of litters per year and survival of pigs born alive due to changes in Turopolje pig production system and development of a health monitoring program.

Keywords. Turopolje pig – Conservation *in situ* – Litter size – Brucellosis.

La taille des portées et la gestion de la santé comme facteurs limitants de la conservation "in situ" du porc Turopolje

Résumé. *Depuis 1996, le porc Turopolje, inclus dans un programme de préservation «in situ», est reconnu comme une race autochtone extensive en Croatie. Le présent travail a pour objectif d'analyser les variations de la population reproductrice de porc Turopolje en rapport avec les caractéristiques reproductives et l'épidémie de brucellose. On a analysé 529 portées de 388 truies dans une période comprise entre 2000 et 2009. On a observé 1,2 portée/truie/année en moyenne avec un résultat de 4,21 et 3,90 porcelets nés vivants et sevrés par portée. Le taux de mortalité au cours de la période de sevrage était de près de 10%. En ce qui concerne le nombre de truies, la population reproductrice s'est accrue plus de dix fois au cours de la période de 1996 à 2009, et cinq fois concernant le nombre de verrats. En 2008, le nombre de truies et de verrats a décru d'environ 21% par rapport à l'an 2000, en raison principalement d'une épidémie de brucellose. En moyenne, 44,2% et 30,1% des porcs analysés au cours des années 2008 et 2009 avaient été sérologiquement positifs à l'épreuve de l'infection à* Brucella suis*, et ont été exclus de la population. Ces résultats semblent indiquer l'exigence d'augmentation du nombre de portées par an et de survie des porcs nés vivants en raison des changements dans le système de production du porc Turopolje et du développement d'un programme de surveillance de la santé.*

Mots clés. *Porc Turopolje – Conservation* in situ *–Taille de la portée – Brucellose.*

I – Introduction

Turopolje pig is local pig breed well adapted to continental climate and extensive farming in periodically flooded forests of English oaks and marsh meadows of Turopolje region in central Croatia (Dikic *et al*., 2002; Dikic *et al*., 2006). Since 1996, it was included in a program of conservation in its natural habitat according to old traditional Croatian farming practices (Robic *et al*., 1996). Typically, pigs are kept outdoor during all phases of production cycles with extensive using of woodland (>3 ha/pig). Housing is based on natural covering except during the farrowing and suckling period in which sows and piglets are kept in primitive woodenly buildings type “stanci” with restricted moving area in wood environment. Feeding is based on utilization of natural resources such as pasture, acorn and soil with addition of about 0.5 kg of corn seed per pig, especially in suckling and late fattening period as well in winter season with snow (Juric *et al*., 1997). Reproductive efficiency and growth performance could be highly influenced by weather conditions and control of diseases. In the past, outbreaks of some infectious diseases (swine fever, brucellosis, leptospirosis) were reported which strongly influenced the size of breeding population and production level (Cvetnic *et al*., 2002; Cvetnic *et al*., 2003; Roic *et al*., 2007). The aim of this paper was to analyze the reproductive characteristics of Turopolje pig and changes in breeding population in the period from 1996 to 2009 in relation to brucellosis outbreaks.

II – Materials and methods

1. The size of breeding population

The size of breeding population (number of sows and boars, gilts and young boars) and number of farms were analyzed in period between 1996 and 2009 using a data of “Annual reports - Pig breeding” of Croatian Livestock Center (CLC, 1997 to 2010). The effective size of population (Ne) was calculated according to formula by Falconer and Mc Key (1996):

$$Ne = 4\ x\ \frac{Nm\ x\ Nf}{Nm + Nf},$$

where Nm is number of male (boars) individuals and Nf is number of female (sows) individuals

2. Reproductive traits

Data were collected in the period from 2000 to 2009, in several farms, representing two main farming areas of Turopolje pig: "Turopolje field" – woodland area near the capital Zagreb and “Natural Park - Lonjsko Polje”. The data from 529 litters and 388 sows registered in Herdbook of Turopolje pig breed were collected. At all farms sows were kept under similar conditions in outdoor production system with extensive using of woodland and pasture. Data collected include: farowing index, number of piglets (total born, born alive and weaned piglets) per litter and per sows.

3. Blood sampling and serological tests

Blood samples were collected by venipuncture from vena cava cranialis, vena jugularis or the ear vein. During the year 2008 and 2009 blood samples from 385 and 83 animals were collected, respectively and were serologically tested to *Brucella suis* infection. The Rose Bengal Test (RBT) were used as a screening method and the Complement Fixation Test (CFT) and the indirect Enzyme-Linked Immunosorbent Assay (ELISA) as a "second line" test to confirm the swine brucelossis. All serologicall tests were performed according to World Orgnisation for Animal Health standard procedures (OIE, 2004). The results of the each tests were interpreted according to producer‘s recommendations. Individual results were considered positive when a positive RBT was confirmed either by a positive iELISA or CFT result. A positive result in the RBT, associated with doubtful reactions in either iELISA or CFT was considered as "suspect".

III – Results and discussion

The program of *in situ* conservation of Turopolje pig breed was started in 1996 by opening of the herdbook at Croatian Livestock Center (CLC) by the public association "Universitas Commnitas Nobilium Campi Turopolia" (UCNCT, land community established in 13th century). At the beginning only 12 sows and 3 boars were registered in the herdbook. In the next three years (Table 1) population was increased and number of breeding animals was consolidated at 36 sows and 6 boars in 1999, what could be used as a basic year for the considerable increase in number of breeding population. In the period from 1999 to 2008 the size of breeding population was increased near the 5 times in number of sows and 6 times in number of boars registered in the herdbook. The increase in number of boars and sows resulted in the increase size of effective population, from 20.6 to 98.6 in 1999 and 2007, respectively. According to the FAO criteria for determining breeds at risk (Loftus and Scherf, 1993), Turopolje pig was in critical status from 1996 to 2003 and in the status of endangerment from 2003 to 2008. But in 2008 the numbers of sows and boars was decreased about 21% in relation to 2007 and did not significantly change during the year 2009.

Table 1. The size of breeding population of Turopolje pig in Croatia†

Year	Sows	Boars	Ne††	Gilts	Young boars	Farms
1996	12	3	9.6	-	-	-
1997	17	8	21.8	-	-	-
1998	13	6	16.4	-	-	-
1999	36	6	20.6	13	6	5
2000	40	5	18.8	37	1	5
2001	45	5	18.0	90	1	4
2002	70	4	15.1	53	3	3
2003	99	6	22.6	76	3	2
2004	116	9	33.4	78	8	2
2005	129	14	50.5	107	-	3
2006	137	13	47.5	8	3	4
2007	164	29	98.6	2	2	4
2008	130	15	53.8	21	7	5
2009	138	18	63.7	83	-	5

†Source: Annual report – pig breeding, HPA (1997 - 2009). ††Ne – effective size of population.

Slow increase of breeding population could be explained due to low reproduction efficiency (low farrowing index, litter size), high mortality during suckling period, appearance of some diseases and low interest of family farms (low daily gain, low meatiness) as well as the lack of pastures and forests areas for pig keeping which are required for *in situ* conservation (Dikic *et al*., 2008).

As shown in Table 2, low farrowing index (in average 1.19) and high variability in number of total born piglets per litter and sow (3.21 to 5.68 and 3.21 to 7.40 piglets per litter and sow, respectively) were observed. In the same time more than 90% of total born piglets were born alive, while mortality during suckling period varied between 2% and 35%. Similar mortality rate was founded by Đikic *et al*. (2008) in semi-controlled conditions up to 21st day (8.8%) and 42nd day (26.5%) of piglets age. The exception was year 2004, when was observed 2.0 litters per sow, but only 3.29 total born piglets/litter and 80% of mortality during suckling period. These results suggest that *in situ* conservation of Turopolje pig in outdoor extensive production system is more dependent on environmental conditions (weather conditions, keeping area and availability of natural feed resources –growing plants, forest fruits, and soil animals– and that one farrowing per year in warm season with abundance of vegetation is sufficient. On the contrary, if the practice of two farrowing per year is used, one of these comes in the cold and humid season, what could have a strong influence on survival of young piglets.

Table 2. Reproduction traits of Turopolje pig breed

Year	Number of litters	Farrowing index	Number of piglets					
			Average per sow			Average per litter		
			Total born	Born alive	Weaned	Total born	Born alive	Weaned
2000	37	1.12	6.24	6.15	4.85	5.57	5.49	4.32
2001	23	1.10	6.14	5.95	3.90	5.61	5.43	3.57
2002	11	1.00	-	-	4.55	-	-	4.09
2003	36	1.00	4.78	3.33	2.83	4.78	3.33	2.83
2004	120	2.00	6.57	6.57	1.32	3.29	3.28	0.66
2005	39	1.00	3.21	3.13	3.08	3.21	3.13	3.08
2006	20	1.33	7.40	7.40	7.13	5.55	5.55	5.35
2007	34	1.17	6.66	6.48	6.41	5.68	5.53	5.47
2008	29	1.16	5.96	5.88	5.32	5.14	5.07	4.59
2009	60	1.02	5.64	5.34	5.15	5.55	5.25	5.07
Total	**529**	**1.19**	**5.26**	**5.02**	**4.45**	**4.44**	**4.21**	**3.90**

The breeding population of Turopolje pig is owned by two public organizations (UCNCT and Lonjsko Polje Nature Park) with more than 50 reproducers each and several small family farms with 3 to 10 animals each in relatively closed breeding area. Such farm structure and production system contributes to high exposure of Turopolje pig population to appearance of infection diseases. In the past, the contagious diseases like leptospirosis, swine fever and brucellosis were reported in pigs and/or in wild animals in the breeding area of Turopolje pig (Cvetnic *et al*., 2002; Roic *et al*., 2007), what emphasize problems in control of infectious diseases in outdoor kept Turopolje pig. Recently, the most important problem are brucellosis outbreaks in several herds of Turopolje pig.

As shown in Table 3, in the years 2008 and 2009, about 44% and 30% of tested blood samples of Turopolje pigs were serologically positive to *B. suis* infection, respectively.

Table 3. Seroprevalence of brucellosis in Turopolje pig breed in year 2008 and 2009

Year	Total number of blood samples	Serological positive blood samples	
		Total	%
2008	385	170	44.16
2009	83	25	30.12
Total	468	195	41.67

Swine brucellosis is a contagious disease in pigs caused by the bacteria *Brucella suis* that could be transmitted to humans. Infection occur primary in domestic and feral pigs, while wild pig (*Sus scrofa*) and/or European hare (*Lepus europeus*) could be assumed as a natural reservoir of *B. suis*. The most common symptoms of brucellosis include non-specific reproductive disorders such as infertility, abortion at any stage of gestation, stillbirth, weak pigs and orhitis in boars. The disease spreads by semen during coitus and by ingestion or inhalation of bacteria in reproductive fluids, placenta, aborted fetuses, urine or milk. Pigs kept in extensive outdoor production systems such as Turopolje pigs are highly exposed to B. suis infection. Pigs often graze together with other species of domestic animals and mate naturally. All of this increases the possibility of direct contact among pigs from different owners as well as with wild boar (Cvetnic *et al*., 2003). In addition, an earlier study (Cvetnic *et al*., 2009) suggests that brucellosis is enzootic in Croatian populations of wild pigs, especially in the breeding area of Turopolje pig. Thus, the presence of *B. suis* in pigs environment and the extensive production system could explain the occurrence and high prevalence of brucellosis in Turopolje pig population.

According to Croatian legislation, all serologically positive animals to *B. suis* infections must be excluded from population. As a result, a lot of animals were excluded from breeding population what could explain the decrease in number of breeding animals in the Herdbook in 2008 without significant changes in number of breeding animals in 2009.

IV – Conclusions

In situ conservation of Turopolje pig breed is based on outdoor keeping according to a traditional production system in which large areas of pasture and woodland are used (more than 3 ha/pig) with low investment in housing and feeding. The main limiting factors for any considerable increase in size of breeding population in last 10 years were: reproduction efficiency (low farowing index, small litter size, high mortality of piglets in suckling period and consequently low number of weaned piglets per sow per year), the size and number of breeding farms and the outbreaks of infections diseases (brucellosis, swine fever, leptospirosis). In order to change the present state the urgent development and implementation of herd health management program and improvements in production system (housing and feed supplementation in suckling period) are needed.

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Pathogenic bacterial contamination of carcasses from outdoor organically reared pigs: preliminary results from a survey in Northern Italy

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Abstract. Organic pigs often are reared and fattened outdoor; this production system normally affects skin cleanliness especially during wet season. The slaughtering of very dirty pigs may increase the risk of carcass contamination. The following survey investigates the presence of several pathogenic bacterial for human foodborne such as *Listeria monocitogenes*, *Salmonella* spp., *Escherichia coli* O157, *Yersinia enterocolitica* and *Campylobacter* spp. Different samples (skin swab sponges performed before and after slaughtering, faeces, tonsils and rind) from risky areas for contamination have been collected from 31 fattened pigs coming from 7 organic farms and slaughtered in 3 small abattoirs. None of those samples were positive for *Salmonella* spp. *Campylobacter* spp., was found on 64.5 % of samples (77% of samples were positive for *Campylobacter coli*). *Listeria monocytogenes* and *Yersinia enterocolitica* were detected on 13% and 6.5% of samples respectively; *Escherichia coli* O157 was found only on 3.2% of considered samples. Preliminary results on the level of carcass contamination show similar figures as found in literature for *Campylobacter* spp., *Listeria monocitogenes* and *Yersinia enterocolitica*, both for the outdoor organic pigs and the conventional ones. The high presence of *Campylobacter* spp. in faeces suggests to pay particular attention during evisceration to prevent carcass contamination.

Keywords. Pig – Organic – Pathogens – Carcass.

Contamination de carcasses de porcs élevés en système biologique en plein air par des bactéries pathogènes: résultats préliminaires d'une étude réalisée en Italie du Nord

Résumé. *Souvent élevés en plein air, les porcs bio, au moment de l'abattage, en particulier pendant la saison des pluies, se présentent très sales, ce qui peut représenter un risque accru de contamination de la carcasse. Dans cette étude a été évaluée la présence de certains pathogènes périlleux pour la consommation de viande:* Listeria monocitogenes, Salmonella *spp.,* Escherichia coli *O157,* Yersinia enterocolitica *et* Campylobacter *spp. Les matrices qui ont été soumises à l'analyse (tampons cutanés avant et après l'abattage, fèces, amygdales et couenne) ont été prélevées sur 31 porcs provenant de 3 abattoirs et élevés dans 7 fermes. Les données obtenues ont démontré l'absence de* Salmonella *spp. dans tous les échantillons. La présence de* Campylobacter *spp. a été relevée dans 64,5%, avec une prévalence de* Campylobacter coli *(77%). 13% des porcs se sont avérés positifs pour* Listeria monocytogenes*;* Yersinia enterocolitica *était présente dans 6,5% des sujets analysés; pour les deux la couenne a été la matrice la plus contaminée.* Escherichia coli *O157 a été relevée seulement dans 3,2% des échantillons. Les niveaux de contamination obtenus pour* Campylobacter *spp.,* Listeria monocitogenes *et* Yersinia enterocolitica *se sont montrés compatibles avec les données citées dans la littérature par rapport aux fermes conventionnelles. La présence consistante de* Campylobacter *spp. dans les fèces nous impose de prêter une attention particulière à l'éviscération, en tant que facteur de risque de contamination des carcasses.*

Mots-clés. Porc – Biologique – Pathogènes – Carcasse.

I – Introduction

Organically reared pigs in Italy has been growing during the last five years due to the increasing of consumer demand for organic products, and for a regional plans that support the use of local-autochthonous pig breeds. However, in Veneto Region (north-east of Italy) organic pig farms are still limited in number and small in size (about one hundred fattened pigs/year/farm), if compared with conventional ones. Differently from conventional, organic pigs are often reared in outdoor systems, using grazing areas with different type of soil, cover crops (grass, corn, sorghum, etc.) or bushes, and slaughtered mainly during wet and cool season. At slaughtering normally pigs are quite dirty and skin is often covered with mud and faeces; this may increase the risk of carcass contamination especially when slaughtering is performed in a small abattoir, often used in local organic production chain. The presence of foodborne pathogens, as well as the related contamination of carcasses and processing facilities, could dramatically increase the health risk, especially if carcasses are deboned and processed at farm level for traditional fermented salami production. Representing a high risk for human foodborne diseases, meat contamination from pathogenic bacteria such as *Listeria monocitogenes, Salmonella* spp., *Escherichia coli* O157, *Yersinia enterocolitica* and *Campylobacter* spp. are under continuous investigation by local health authorities.

A previous investigation carried out on the same area (Schiavon, *et al*., 2006) has shown a similar (*Campylobacter* spp.) or higher (*Yersinia enterocolitica*) contamination of outdoor organic pork carcasses compared to the conventional ones. In this second investigation different farms and small abattoirs were included, with the aim of improve the knowledge on the presence of critical foodborne pathogens, in order to suggest different or more careful management of organic outdoor pigs.

II – Materials and methods

1. Sampling

The research has been carried out by sampling 31 pigs produced in 7 outdoor organic farms located in the Veneto region (plane and low mountain areas); animals have been randomly chosen at farm level and slaughtered in 4 different abattoirs, during winter season (from November to February).

Five samples have been collected from each animal: skin swab before slaughtering, skin swab post-slaughtering (at the end of slaughtering line before carcass cooling), faeces, tonsils and a portion of rind (a strip about 5 x 15 cm length on belly and thorax area).

The skin swabs before and after slaughtering has been performed through sponges BS10-BPW (Qualicum Scientific Limited) on dorsal, flank and jaw regions, using one sponge on the three regions.

All samples has been microbiologically investigated for presence of *Salmonella* spp., *Yersinia enterocolitica*, *Listeria monocitogenes*, *Campylobacter* spp. and *Escherichia coli* O157, apart tonsil that were investigated for *Yersinia enterocolitica* only.

2. Analytical methods

***Salmonella* spp.:** Samples were suspended in buffered peptone water (ratio 1:10), incubated overnight at 37°C, then inoculated on MRSV (semisolid) medium and incubated overnight at 42°C. Positive samples were streaked into BGA and XLD media and incubated at 37°C overnight. In case of suspect, colonies were identified through biochemical reaction and confirmed with serotyping.

***Campylobacter* spp.:** Samples were suspended in Preston broth (ratio 1:10) and incubated overnight at 42°C. Then they were streaked on Karmali agar plates and incubated at 42°C for 24-72 under modified air. After Gram staining, the suspected colonies have been identified through PCR.

***Yersinia* spp.:** Samples were homogenized in bile-sorbitole broth (ratio 1:10) and incubated at 4 °C for 10 days. After this, the samples were streaked into selective CIN agar and incubated at

37°C overnight; suspected colonies were identified by biochemical methods and confirmed using commercial API 20E system (BioMérieux Italia SpA).

***Listeria* spp.:** Samples were homogenated with Half Fraser Broth (ratio 1:10) and streaked on agar blood plate and were incubated for 24 hs at 37°C. From HFB samples were inoculated into solid media (Oxford agar and Aloa agar) incubated 48 hs at 37 °C and suspected colonies were identified using biochemical method (API test).

***E. coli* O157:** Samples were suspended in Triptone soy broth (ratio 1:10) and incubated overnight at 41,5°C. After this, *E. coli* O157 is concentrated using magnetic immunoseparation method, streaked on sorbitol McConkey agar and incubated at 37°C overnight, and confirmed using biochemical and serological methods.

III – Results

All data are shown in Tables 1 and 2.

Table 1. Presence of pathogens in samples: % prevalence and absolute numbers

Pathogen samples	*Salmonella* spp.	*Listeria monocitogenes*	*Yersinia enterocolitica*	*Campylobacter* spp.	*Escherichia coli* O157
Pre-slaughtering swab	0	6.5 (2/31)	0	9.7 (3/31)	0
Post-slaughtering swab	0	3.2 (1/31)	0	9.7 (3/31)	0
Pork-skin	0	9.7(3/31)	6.5 (2/31)	12.9 (4/31)	0
Tonsils	-	-	3.2 (1/31)	-	-
Faeces	0	0	0	51.6 (16/31)	3.2 (1/31)

Table 2. Presence of pathogens in samples of each farm

Farm	Pigs tested (n)	*Salmonella* spp.	*Listeria monocitogenes*	*Yersinia enterocolitica*	*Campylobacter* spp.	*Escherichia coli* O157
1	7	neg	pos	neg	pos	neg
2	5	neg	neg	neg	pos	neg
3	6	neg	pos	neg	pos	pos
4	6	neg	neg	neg	pos	neg
5	1	neg	neg	neg	neg	neg
6	2	neg	neg	neg	pos	neg
7	4	neg	neg	pos	pos	neg

None of the 31 pigs sampled has shown the presence of *Salmonella* spp.

Campylobacter spp, was found in 20 animals (64,5%) with higher prevalence on faeces as expected from previous experiences. On 26 strain isolated, 20 were identified by PCR as *C. coli* and 6 as *C. jejun*i.

Listeria monocitogenes was found in 4 pigs (13%), only on skin.

Yersinia enterocolitica was found only in 2 pigs (6,5%); one presented tonsils (both) and skin contaminated, the other one the skin only.

Escherichia coli O157 was found only on one pig's faeces (3,2%).

IV – Discussion

The analysis of the data underlines a possible unfriendly situation, depending from the bacterial strains under investigation.

No *Salmonella* strains were isolated, but the low number of samples analized could not guarantee the absence of *Salmonella* in the farms. These findings may suggest that the bio-safety level could be considered quite good on the investigated farms. However, it seems reasonable that in outdoor organic pig production, the source of possible contamination of *Salmonella* has to be carefully investigated, in relation to rearing system and the possibility of contact between pigs and wild animals.

Listeria monocytogenes was found in 13% of the slaughtered pigs, but only on their skin; moreover only pigs from two farms were contaminated. This could suggest a cross-contamination between pigs and ground. According to some literature findings, our data confirm that *Listeria monocytogenes* derive more from the environment (farm or processing environment) than from the animals (Samelis and Metaxopoulos, 1999). This confirms the importance of monitoring the presence of *L. monocytogenes* in the environment where the pigs live, on transport vehicles, on abattoir lairage and on processing plant.

Campylobacter spp. is the most common pathogen isolated. It was found in 64.5% of the slaughtered pigs coming from 6 farms. According to literature and as for conventional pigs, its prevalence is higher in faeces (51,6%) than on skin, and *C. coli* prevalence is higher compare to *C. jejuni* (Schiavon, *et al*., 2006; Farzan *et al.*, 2010). It seems enough clear that to lower as much as possible the prevalence of Campylobacter spp. on pig carcasses, the main critical point should be to guarantee a higher level of hygiene (expertise of operators, knives sterilization, etc.) during the evisceration process.

The prevalence of *Yersinia enterocolitica* is very low, and similar to as reported in literature for wild boar. Only pigs coming from one farm were contaminated. Tonsils, known as a target organ for this bacteria, present a very low contamination level. However, a careful processing technique has to be implemented in order to minimize the risk of cross contamination during processing and foodborne associated to pork consumption.

The prevalence of *E. coli* O157 is very low, too; it was found into the faeces (on one pig only) similar to investigation reported by Ercolini *et al*.,(2007) for wild boar.

Our findings confirm that the risk associated with carcasses from outdoor organic pigs are similar to which of intensive reared pigs. The most prevalent foodborne pathogen was *Campylobacter coli*, with less prevalence of *Campylobacter jejuni* and *Listeria monocytogenes. Yersinia enterocolitica* and *E. coli* O157 were very low in prevalence, and *Salmonella* spp. was absent.

Related risk can be minimized using appropriate processing methods; furthermore it is important to investigate foodborne risk associated to outdoor organic reared pigs, as the market of this product is increasing. The low level of main investigated foodborne pathogens, in the majority of farms, indicates that it could be possible to obtain safe pig carcasses from controlled outdoor organic production system.

Acknowledgements

The authors wish to acknowledge the financial support received from the Veneto Region (Project BIOVENETO Biodemo_azione suini).

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KRL test to objective evaluation of welfare: sensibility to housing conditions and dietary supplements

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Abstract. The aim of this work was to determine the sensibility of KRL test to evaluate pig welfare in relation to different housing conditions and dietary supplements. It is reported that the response to oxidative stress could be considered as welfare parameter in swine. The KRL test allows the evaluation of total blood antioxidant activity. In the first trial we evaluated the total blood antioxidant activity of 12 swine allotted respectively to solid and totally slatted floor. In the second trial blood samples were obtained at 0, 15 and 60 days from 10 weaned piglets fed control diet and diet supplemented with vitamin E (54 mg/kg) respectively. The first trial showed that the total antioxidant activity of red blood cells resulted higher in pig allotted to the solid floor (P=0.01). In the second trial the KRL test evidenced an increase in the total antioxidant activity of whole blood and red blood cells of piglets fed diet supplemented with vitamin E ($P<0.05$). The present research showed that the KRL test is able to discriminate welfare in relation to different housing conditions and dietary supplements.

Keywords. Total blood antioxidant activity – KRL test – Welfare – Pig.

Emploi du test KRL pour une évaluation objective du bien-être : sensibilité aux systèmes d'élevage et à la supplémentation alimentaire

Résumé. *La sensibilité du test KRL pour l'évaluation du bien-être des porcs a été étudiée en relation avec le système d'élevage et la supplémentation alimentaire. La réponse au stress oxydatif est un paramètre pour l'évaluation du bien-être chez le porc. Le test KRL permet de mesurer la résistance globale des individus vis-à-vis de l'agression des radicaux libres. Dans un essai d'engraissement le pouvoir de défense antiradicalaire global a été évalué sur 12 porcs élevés sur béton plein ou sur caillebotis intégral. Dans le second essai des prélèvements sanguins sont réalisés au sevrage, à 15 et 60 jours après sevrage sur 10 porcelets allotés en 2 régimes alimentaires identiques, à l'exception de l'ajout de vitamine E (54 mg/kg). L'activité antioxydante totale des hématies est plus élevée chez les porcs élevés sur béton plein (P=0,010). Le test KRL démontre une augmentation du pouvoir de défense antiradicalaire global du sang et des hématies chez les porcelets alimentés avec vitamine E (P<0,05). En conclusion, le test KRL est capable de discriminer le bien-être en relation avec le système d'élevage et la supplémentation alimentaire*.

Mots-clés. *Pouvoir de défense antiradicalaire global – Test KRL – Bien-être – Porc.*

I – Introduction

The management of production animals has changed radically across the European Union (EU) over the past five decades. Over this time, animal agriculture has intensified and the animals have been moved to indoor housing systems with higher stocking densities. Over the past years a considerable amount of scientific research has focused on animal welfare (Broom 1991; Sandøe *et al.*, 2003). Performance records, behavioural, physiological and clinical parameters are consider as good indicators for assessing animal welfare (McGlone, 2001; Broom 1996). The welfare in pig specie could be evaluated through three different basic approaches. The first method is based on the normal biological functioning including the physical and physiological condition of the animal. The second approach regards animal feelings (Broom 1991) and it can be assessed using ethological parameters. The third, calls functional approach, is based on physiological, immunological and pathological indicators, that currently offers interesting data on pig welfare (Barnett at al., 2001).

In the recent years oxidative stress became an important goal in human and animal research as cause of some disease. The "reactive oxygen metabolites" (ROM) are produced endogenously by normal metabolic processes, but amounts may be increased markedly by exogenous factors (Machlin and Bendich 1987). Deficiencies of natural protective substances or excess exposure to stimulators of ROM production may result in oxidative stress, defined as an imbalance between oxidants and antioxidants at the cellular or individual level (Finkel and Holbrook, 2000).

In farm animals, oxidative stress may be involved in several pathological conditions, that are relevant for animal production and the general welfare of the individuals (Lykkesfeldt *et al.*, 2007). Diseases such as pneumonia and sepsis have been shown to involve altered redox balance in pigs (Lauritzen *et al.*, 2003; Basu and Eriksson, 2000). Brambilla *et al.*, (2002) reported that the response to oxidative stress can be utilize as welfare parameters in pigs.

Methods for quantifying oxidative stress include the assessment of the total antioxidant capacity of serum or plasma. The Trolox equivalent antioxidant capacity (TEAC) assay, the oxygen radical absorbance capacity (ORAC) assay, and the ferric reducing ability of plasma (FRAP) assay are commonly used and have been extensively evaluated. These methods allow to measure the total antioxidant capacity of serum or plasma without consider the antioxidant defences in the red blood cells.

The KRL test shows the resistance to free radicals assessed as the time needed to haemolyse 50% of red blood cells exposed to a controlled free radical attack and provides an assessment of total antioxidant defences, since all families of antioxidants present in whole blood are used to fight off the oxidant attack (Stocker *et al.*, 2003; Girard *et al.*, 2005). The KRL test has several applications*;* in humans KRL is used to study the effectiveness of natural or pharmaceutical treatments and to discriminate welfare conditions depending on medium or high stress.

In the pig production chain, the relationships between housing system and welfare became an important goal of interest (EFSA 2005). The floor type is a determinant factor of certain foot lesions (Gillman *et al.*, 2009) and can affect health, performance and behaviour (Ruiterkamp 1987). In particular on totally slatted floor the animal movement decreases and they spend more time lying down (Rossi *et al.*, 2008). Higher stress during pre-slaughter handling was also observed in pigs reared on totally slatted floor than on solid floor (Nanni Costa *et al.*, 2007).

Another goal of interest are the elevate oxidative stress in the post weaning phase that can lead to increased prevalence of infectious disease. Weaning are reported as one of the critical stages for dietary vitamin E as a nutrient for growth and health status in pigs (Lauridsen *et al.*, 2005). In fact the weaning-induced stress and the decline in the antioxidant level causes an elevated oxidative status in weaned piglets (Rossi *et al.*, 2009; Sauerwein *et al.*, 2005).

The goal of the present experimental work was to determine the sensibility of KRL test to evaluate pig welfare in relation to different housing conditions and dietary supplements.

II – Materials and methods

Two independent trials were performed to test the KRL test sensibility to discriminate the effect of the housing conditions and dietary supplement on total blood antioxidant activity in pigs.

In the first trial 24 barrows of a live weight in the range of 80 to 120 kg, half allotted to solid floor and half to totally slatted floor, were randomly selected. Fasting blood samples were taken by anterior vena cava puncture, collected in 10 mL vacutainer glass tubes containing EDTA (Venoject®, Terumo Europe N.V., Leuven, Belgium) and immediately placed on ice pending analysis. The analysis were performed within 24 h from the sampling procedure.

In the second trial eighty weaned Dalland piglets of an average weight of 7 kg, were assigned, on the basis of weight and sex, to two dietary treatments: control diet (175 mg/kg of vitamin E) and a diet supplemented with the basal level plus 54 mg/kg of vitamin E (225 mg/kg of vitamin E). The experimental diets were formulated to be isoenergetic and to meet or exceed the NRC requirements for all nutrients, and was presented for *ad libitum* consumption. At weaning, 15 and 60 d post weaning, 10 piglets per treatment were randomly selected and fasting blood samples were taken by anterior vena cava puncture, collected in 10 ml vacutainer glass tubes containing EDTA (Venoject®, Terumo Europe N.V., Leuven, Belgium) and immediately placed on ice pending analysis. The analysis were performed within 24 h of collection.

Total antiradical activity of whole blood and red blood cells (RBC) for each pig was evaluated using KRL biological test (Laboratoires Spiral, France). The KRL test is currently used to test the capability of erythrocytes to resist a standardized production of free radicals generated from the thermal decomposition of a 27 mmol/L solution of 2,2′-azobis (2-amidinopropane) hydrochloride at 37 °C (Prost, 1992; Blache and Prost, 1992). Whole Blood and RBC samples diluted to 1/50 were submitted in isotonic saline solution to organic free radicals. Haemolysis was recorded using a 96-well microplate reader by measuring the optical density decay at 450 nm (Fig. 1).

Fig. 1. KRL reader and 96-well microplate.

Results were expressed as the time required to reach 50% of maximal haemolysis (half-haemolysis time - HT_{50} - in minutes), which refers to the whole blood resistance to free-radical attack.

In the first experimental trial one-way ANOVA was used to determine statistically significant differences between the two housing conditions. In the second experimental trial the data were analyzed used a repeated measure ANOVA with the weaning value entered as a covariate. Data are presented as mean ± SEM. Differences between means were considered significant at $P<0.05$.

III – Results and discussion

In the first trial no differences in the total blood antioxidant activity were observed in pigs reared on the two different kinds of floor (P = 0.320) (Figure 2A). Considering the antioxidant activity of the RBC, an higher value was observed in pigs reared on solid floor that on totally slatted floor (P = 0.010) (Fig. 2B).

The RBC value concerns especially the intracellular defence status and it is important for understanding the balance between attack and defence of organism in a medium/long period, considering that RBC mean life in pig is 60-85 days (Pastorelli *et al.*, 2009). This result allows to hypothesize that the environmental stress, due to the totally slatted floor, cause an intracellular imbalance between the oxidative and antioxidant systems that make the cells more susceptible to oxidative damage (Brambilla *et al.*, 2002). The above findings point out that the adaptive response to a long term stressing condition implies an impairment of this reaction. During the state of chronic stress negative effects on health and the development of pathological conditions involving oxidative stress may occur (Lykkesfeldt *et al.*, 2007).

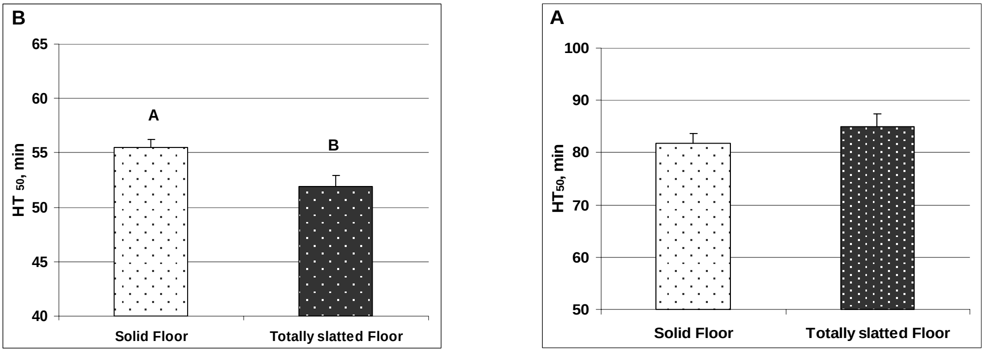

Fig. 2. Whole blood (A) and red blood cells (B) antioxidant activity in pigs reared on solid or totally slatted floor. Data are reported as means ± SEM; n=12; A, B for P = 0.010.

The second trial regards the KRL test sensibility to the dietary supplementation of vitamin E in post weaning piglets. The results show that the total antioxidant activity in both whole blood and RBC was enhanced (P < 0.05) by a supplemental dosage of dietary vitamin E (Fig. 3).

A higher supplementation with vitamin E in the post weaning phase improves the total blood antioxidant activity, according with an our previous study that reported a decrease in the ROS production after dietary supplementation of natural antioxidant (Corino *et al.*, 2007). These data give clear evidence that dietary antioxidant supplementation after weaning may positively affect the antioxidant status, and therefore improving pig health.

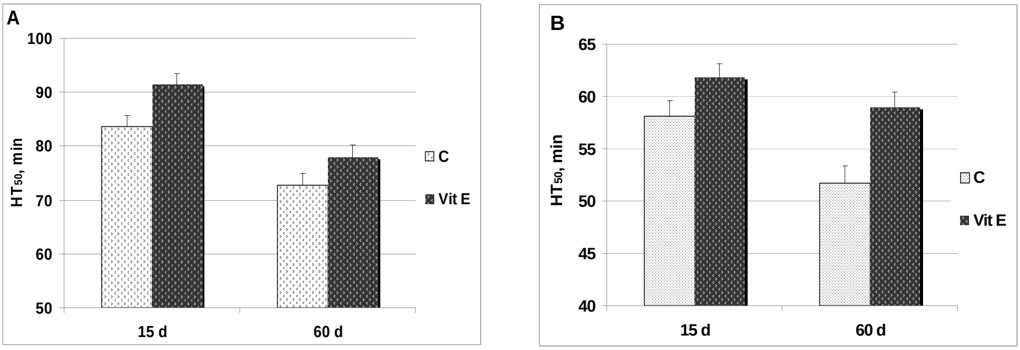

Fig. 3. Whole blood (A) and red blood cells (B) antioxidant activity in piglet fed control or vitamin E supplemented diet. Data are reported as means ± SEM; n=10; C and Vit E treatments differ for P<0.05.

IV – Conclusions

Under the conditions of the present research, the KRL test is able to discriminate pig welfare in relation to different housing conditions and dietary supplements. The housing system with totally slatted floor negatively influenced RBC antioxidant activity, evidencing a sign of a chronic stress. The supplemental dosage of vitamin E are able to increase blood total antioxidant activity, suggesting a better resistance to oxidative stress occurring in the post weaning phase. These results confirm that the KRL test should be consider as a new laboratory analysis to assess welfare in pig specie.

Acknowledgements

This work belongs to a research project called "Nuove metodologie per la valutazione oggettiva del benessere animale nella specie suina" financed by Regione Lombardia – Agricultural Department, according to the Plan of Research and Development "2008".

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Session 5
Meat quality and products

Quality of meat and cured products of Mediterranean autochthonous pigs

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Abstract. Meat quality characteristics of Mediterranean pigs as affected by genetic and environmental effect are summarised and discussed. From the recent literature it is evident that the Mediterranean local breeds have interesting quality of meat and fat respect to the improves one. The link with free-range rearing increases the commercial value of products of local pigs, because of both effective characterization and consumer suggestion, research is helpful to increase the knowledge of their rearing system.

Keywords. Mediterranean autochthonous pigs – Meat quality – Free range – Dry-cured products.

Qualité de la viande des produits secs des porcs autochtones méditerranéens

Résumé. *Les effets du type génétique et du système d'élevage sur les caractéristiques de la qualité de la viande et des produits secs des races porcines autochtones méditerranéennes ont été examinés. La littérature récente montre que les races porcines ont des paramètres qualitatifs de la viande et de la graisse qui sont souvent supérieurs à ceux des porcs améliorés. Le lien avec le troupeau sauvage augmente la valeur commerciale des produits du porc local, en raison de la forte caractérisation et de la suggestion du consommateur qui en résulte.*

Mots-clés. *Races porcines indigènes méditerranéennes – Qualité de la viande et des produits secs – En plein-air.*

I – Introduction

In the countries of the Mediterranean area there is a increasing interest for autochthonous pig as the numerous scientific trials reviewed in this paper demonstrate. In the web site of FAO (http://dad.fao.org/), is shown the list of local breeds in the main Mediterranean countries.

1. The main local pigs in the Mediterranean area

In Croatia the main local breed is the Black Slavonian created in the second half of the 19th century. Until the 1950s it was the most widespread breed in the Slavonia, mainly used for the production of fat and meat products. Recently, the population was drastically reduced and in 1990s the survival of the breed was endangered. Due to current protection measures the effective population rather increased; in 2006 there were 46 boars and 604 sows. The breed is well adopted for outdoor keeping (Karolyi *et al*., 2007). In France, at the present, the main local breeds of interest are: the Porc Blanc de l'Ouest, the Porc Limousin, the Porc Gascon, the Porc de Bayeaux, the Porc Basque and the Porc Nustrale (ex Porc Corse).

The conservation of these breeds is intended, as in other European countries, to maintain genetic variation within-breeds and to obtain economics advantages through the obtainment of high quality products. As regard Greek pig, it is one of the oldest Mediterranean pig breeds. Currently its rearing is expanding, due also to public subsidies to the farmers. In the years 70-90s it faced the extinction because of the strong competition of improved genotypes and crossbreeding with wild boars. Although the extinction risk is still considerable, at present the statistics report the presence of approximately 350 sows and a total population of 2000 black pigs. In Italy, among the several local pigs listed by FAO (Table1), only six autochthonous swine breeds are, at the present, well survived to the well-known problems linked to the social-economical transformations of the agricultural world happened in the last century; they are: Apulo-Calabrese, Casertana, Cinta Senese, Mora Romagnola, Nero Siciliano and Sarda.

Table 1. Local breeds of some Mediterranean countries listed by FAO(http://dad.fao.org/).

France	Italy	Spain
Bayeux	Apulo-Calabrese	Celta
Blanc del'Ouest	Bergamasca nera	Chato Murciano
Carélie	Casertana	Gochu Asturcelta
Corse	Cinta Senese	Iberico
Creole	Macchiaiola Maremmana	Ibérico (Dorado Gaditano)
Gallia	Mora Romagnola	Ibérico (Mamellado)
Gasconne	Napoletana Fulva	Ibérico (Negro Entrepelado)
Pie Noir du Pays Basque	Nero dei Lepini	Ibérico (Negro Lampiño)
Porc de Saint Yriex	Nero dei Monti Dauni Meridionali	Iberico (Retinto)
	Nero Reatino	Ibérico (Torbiscal)
	Nero Siciliano	Manchada de Jabugo
	Parmigiana Nera	Negra Canaria
	Pugliese	Negra Mallorquina
	Sarda	
	Siciliano	
	Suino dei Nebrodi e Madonie	
Greece: Greek; **Portugal:** Alentejana and Bisaro; **Slovenia:** Krskopolje; **Croatia:** Black Slavonian and Turopolje		

These breeds have been recognised with the establishment of the Anagraphic Register. The recovery of genetic variability represent an indubitable vantage for breeders; through the valorisation and characterization of autochthonous breeds they can offers at consumers products with high added value. Moreover Italian consumers have an increasing interest toward "niche products". Besides those reported by FAO (Table 1), in Portugal are officially reared three breeds of local pigs: Bisara, Malhado de Alcobaça and Alentejano. In recent years there has been a consolidation and expansion of Portuguese local pig and many changes in production, processing and marketing are taking place. In Slovenia the only autochthonous breed of pig is the Krskopolje, or "blackbelted", who’s origin is the south-eastern part of the Slovene region of Dolenjska. This is an extensive breed, whose characteristics are resistance, good adaptability to poor rearing and feeding conditions and excellent meat quality. In Spain, besides some local breeds are at present reared, the Iberian breed is certainly the most famous pig and it can be considered the best example of the strong cooperation among public institutions, producers and scientific world.

In recent years productive performances of Mediterranean autochthonous breeds have been investigated by several Authors, that take into account many affecting factors. This paper will discuss only the trials that consider the effect of genotype and rearing system being the latter the main factors that affect quality of products of local breeds.

II – Quality of meat and cured products

The Mediterranean pig breeds are reared with different modalities that go from systems that, even if outdoor, foreseen the total food supply with concentrate, to more extensive systems up to the rearing where the fattening phase is carried out using only the spontaneous resources of the wood. The latter is the typical example of the "Montanera" system where the Iberian pig are reared in the "Dehesa" (López-Bote, 1998) and of the "Montados" for local Portuguese pigs (Tirapicos-Nunes, 2007). In the extensive pig production of the Mediterranean area the genotype-environment interactions result in measurable effects on pig meat quality (Edwards and Casabianca, 1996).

1. Effect on quality of lipids

As previously pointed out, the outdoor rearing of local pigs is carried out almost on pasture in forest, so it is very different from the classic outdoor system used for improved pig where feeding is based on commercial feed. Consequently, the effect of food source, especially on the quality of lipids, is very strong (Table 2). As reported by various Authors (Dìaz *et al.*,1996; Coutron-Gambotti *et al.*, 1998; Andrés *et al.*, 2001; Cava *et al.*, 1999a-2000a), the adipose tissue of outdoor-pigs reared in woods is characterised by a high content of unsaturated fatty acids. Coutron-Gambotti *et al*. (1998) found higher percentage of polyunsaturated and monounsaturated fatty acids in Corsican pigs fed chestnuts than commercial diet. In the Iberian pigs fed acorn higher MUFA (primarily oleic acid) and lower SFA contents (primarily palmitic and stearic acid) were found both in intramuscular fat (Cava *et al*., 1999a; Cava *et al*., 2000a; Andrés *et al*., 2001) and in backfat (Díaz *et al*., 1996). This result is consistent with the data on Italian breeds Nero Siciliano (Chiofalo *et al*., 2007) and Cinta Senese (Pugliese *et al*., 2005).

Table 2. Effect of pasture on fatty acid composition

Author	Rearing system	C18:1	C18:2	C18:3
Díaz *et al*., 1996, on Iberico (fresh fat)	Acorn + grass	57.1	9.4	
	Concentrate	47.4	8.3	
Andrés *et al*., 2001, on Iberico (TG of fresh BF)	Acorn + grass	53.4	5.6	0.3
	Concentrate	50.7	6.4	0.3
Cava *et al*., 2000, on Iberico (TG of fresh BF)	Acorn + grass	55.1	5.66	0.68
	Concentrate	51.9	5.02	0.64
Pugliese *et al*., 2005, on Cinta Senese (fresh fat)	Pasture on wood (chestnut + acorn + grass)	52.8	11.6	0.87
	Concentrate	50.3	9.5	0.32
Pugliese *et al*., 2009, on Cinta Senese (dry-cured ham)	Acorn	50.8	12.2	0.8
	Chestnut	48.4	13.6	1.0
	Concentrate	46.2	14.6	0.84
Pérez-Palacios *et al*., 2010, on Iberico (dry-cured ham)	Acorn + grass	53.2	6.96	0.64
	High Oleic Concentrate	48.8	5.75	0.27
		MUFA	**PUFA-n3**	**PUFA -6**
Chiofalo *et al*., 2007, on Nero Siciliano (fresh fat)	Traditional diet	49.1	0.94	9.08
	Concentrate	47.6	0.77	10.62

The latter, in addition, showed an higher PUFA content in outdoor, probably because of the contemporary pasture on chestnuts and acorn during fattening. It is well known that chestnuts have higher contents of polyunsaturated fatty acids in comparison to acorns (Coutron-Gambotti *et al*., 1998; López-Bote, 1998). Also Sirtori *et al.*, (2008) found that the substituting concentrate for chestnut affects significantly the fat quality. Chestnut supply led to more unsaturation level of adipose tissue due to the highest PUFA and MUFA content. Another source of PUFA in free range conditions is the grass that, as reported by Muriel *et al*. (2002), is characterized by high levels of linolenic acid. These authors concluded that free-range leads to increasing levels of total n-3 PUFA in neutral and polar lipids and of individual n-3 PUFA, including EPA and DHA.

The effect of pasture on acorn-wood is not the same of the administration of acorn in confinement rearing system. In fact, Zumbo *et al*. (2007a), on *Longissimus lomborum* of Nero Siciliano pigs, found an higher content of MUFA an lower of PUFA in animals fed acorn respect to pigs fed barley. As regard α- and γ-tocopherols they are provided by the free range system and contribute as antioxidants to prevent lipid oxidation even if it seems that the tocopherol accumulation is not modified by the rearing system but by the type of feeding (Rey *et al*., 2006). On Alentejano pigs Neves *et al*. (2007) found a higher level α-tocopherols in pigs reared extensively respect to those reared in intensive condition.

The effect of the extensive rearing system on fatty acid composition of lipids remain also on seasoned products, as confirmed by a wide literature on several autochthonous breeds. Antequera *et al*. (1992) and Cava *et al*. (2000b) reported, in Iberian ham, high concentration of oleic acid supported by acorn pastures that, together with the typical marbling of the meat, is considered essential for appropriate ripening and flavour development of dry-cured products. In some cases the effect of pasture in wood on fatty acid profile was more strong on dry-cured product than on fresh meat. So, Pérez-Palacios *et al*. (2010) found that feeding acorn respect feedstuff with high oleic determined higher MUFA and PUFA content on intramuscular fat of Iberian dry-cured ham, but no effect on fresh meat of the same animals (Pérez-Palacios, 2009). Similar effect of pasture on wood was found on the fatty acid profile of dry-cured product of Cinta Senese pigs (Pugliese *et al*., 2009) where a higher content of MUFA (primarily oleic acid) in seasoned fat of pastured animals were found. The strong genotype X rearing interaction, and the consequent differences in lipids fatty acid composition, is well exploited for Iberian pig products which are classified according to their fatty acid composition to discriminate the commercial value in three commercial types: Montanera, Recebo and Cebo (López-Bote, 1998).

In this regard, to ensure the consumer against fraud, inspection controls are carried out. They are based upon "on farm" inspector visits and fatty acid analysis done in one fat sample taken from a group of animals from the same producer. That system is very time consuming and expensive, and not objective enough to fulfil the increasing consumer demands from regional, national, and international markets (Garrido and De Pedro-Sanz, 2007). For these reason many researches are carried out to try to found new innovative methods of traceability, such as NIRS technology (Pérez-Marín *et al*., 2009), electronic nose (Garcia *et al*., 2003) and neophytadiene content (Tejeda *et al*., 2001).

2. Effect on volatile compounds and sensorial traits

Many researches were carried out on the effect of rearing system effect on volatile compounds of dry-cured products identified by gas chromatography-mass spectrometry analysis (GC-MS) procedures. The effect of rearing system on Iberian ham volatile compounds has been studied (Carrapiso *et al*., 2003; Cava *et al*., 1999; López *et al*., 1992; Jurado *et al*., 2007; Jurado *et al*., 2009) but contradictory results have been reported. As shown by Jurado *et al*. (2007), there are a lot of factors that could influence volatile compounds in relation to rearing system, such as the concentrate composition and the Montanera feeding period length. They could cause a great heterogeneity in the results.

Up to now a great effect of rearing system on volatile compounds has been found when feeding composition is clearly different. In fresh and in seasoned lard of Nero Siciliano pigs fed acorn (Zumbo *et al*., 2007b) and in salami (Zumbo *et al.*, 2007c) and dry-cured ham (Pugliese *et al.*, 2009) of Cinta Senese pigs fed acorn, a significant effect of feeding regime on volatile compound was found. In the last years further analytical techniques of detection of volatile compounds has been developed. Some powerful odorants found in meat systems exist at concentrations too low to allow their identification by the usual gas chromatography-mass spectrometry (GC-MS) procedures while gas chromatography associated with olfactometry (GC-O) could be a useful tool to identify and characterize the odour-active compounds. GC-O may be used to research aroma differences, because samples with different sensory profiles show differences in their odour-active compounds; in addition, odours detected during GC-O could be related to sensory attributes (Carrapiso *et al*., 2002).

In Iberian ham significant differences in the olfactometric profile between pigs fed acorn or commercial feedstuff were found, even if there was not a single contributor to the two feeding systems odours; odour and aroma differences seem to be caused by differences in the concentration of some odour-active compounds (Carrapiso *et al*., 2002). Garcìa-Gonzales *et al*. (2009) carried out a study on the relationship between odours, flavour sensory attributes and volatile compounds in Iberian hams from diverse geographical origins. Also in other Mediterranean products the olfactometric profile was defined, such as in Serrano ham in Spain (Flores *et al*., 1997).

In order to explain how some chemical characteristics, affected by rearing system, can influence the sensorial traits of dry-cured products, many researches are recently focused on the study of relationships between sensorial traits and lipids composition. Carrapiso *et al*. (2003), with regard to the relationships between sensory profile and subcutaneous fatty acid composition, found that palmitic and oleic acid were the most significantly correlated to the largest number of sensory traits. Moreover, large correlations appeared between stearic and oleic acids and brightness, oiliness, juiciness, sweetness, fat hardness and cured aroma of Iberian dry-cured ham. The effect of rearing system on volatile compounds, above stressed, is reflecting also on sensorial traits being a significant correlations between these traits. As reviewed by Gandemer (2009), positive aroma notes, such as "cured ham", or "aged" aroma notes, have been correlated to either branched aldehydes arising from aminoacids degradation or methylketones arising from lipid oxidation; rancid aroma is correlated to oxidation products, mainly to aldehydes such as nonanal and 2-hexanal which exhibit a strong rancid odour.

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Proteomic profile of dry cured ham adapted to low salt content

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Abstract. Proteomic profiles of Bayonne dry-cured ham with reduced salt content (20% approximatly) was established and compared to normal ones. For each animal, one ham followed the traditional salting procedure (11 to 13 days) while the controlateral was salted for a shorter period (8 to 11 days) according to the weight of hams. The non soluble protein fraction from *Biceps femoris* muscles was studied after 12 months processing. Proteomic analysis (two-dimensional electrophoresis, image analysis and mass spectrometry identification) were performed on the 120 hams. 32 spots were significantly affected by salt; among them, 12 proteins and fragments were identified by MALDI TOF. Eight proteins or fragments belongs to the sarcoplasmic proteins (creatine kinase, glycogen phosphorylase, piroxiredoxine, α crystalline,..) and the 4 remaining were 3 isoforms of actine and one actine fragment. Specific pattern was associated to salt content in Bayonne dry cured ham and we demonstrated that salt modified the solubility properties of the sarcoplasmic proteins during processing.

Keywords. Dry-cured ham – Proteolysis – Proteomic profile – Salt.

Carte protéomique du jambon de Bayonne à teneur réduite en sel

Résumé. *Nous avons établi des cartes protéomiques à partir du jambon de Bayonne fabriqué avec une teneur réduite en chlorure de sodium. Pour ce faire, pour chacun des porcs (n=60), un jambon a été salé normalement, à raison de 11 à 13 jours de salage selon son poids, tandis que son controlatéral était salé pendant un moindre temps (de 8 à 11 jours selon le poids). Nous avons étudié la fraction protéique myofibrillaire des muscles* Biceps femoris *des jambons de Bayonne ayant 12 mois de séchage. Les analyses protéomiques (électrophorèse bidimensionnelle, analyse d'image et identification des spots d'intérêt par spectrométrie de masse) ont été réalisées sur les 120 jambons. Au total 32 spots varient significativement en fonction de la teneur en sel. Parmi eux, 12 protéines et fragments protéiques ont pu être identifiés par MALDI TOF, 8 correspondent à des protéines ou fragments protéiques sarcoplasmiques (créatine kinase, glycogène phosphorylase, péroxyrédoxine, αcrystalline, etc.) et les 4 autres sont des molécules d'actine et un fragment d'actine. Nous avons montré que la teneur en sel dans le jambon de Bayonne est associée à un profil protéomique particulier et qu'elle modifie sensiblement les propriétés de solubilité des protéines sarcoplasmiques.*

Mots-clés. *Jambon sec – Protéolyse – Carte protéomique – Sel.*

I – Introduction

Bayonne ham enjoys EU Protected Geographical Indication (PGI) status. This certification requires professional processors to comply with specifications that provide the consumer with a finished product of optimal quality. The curing technology is based on the addition of salt on the ham internal surface, which acts as a preserving agent but is also responsible for causing physico-chemical and biochemical phenomena that contribute to development of the textural properties. Salt affects muscle proteins by inducing denaturation (Adamsen *et al.,* 2006; Graiver *et al.*, 2006) in which the extent depends on salt concentration (Thorarinsdottir *et al.*, 2002), and processing yields (Santé-Lhoutellier *et al.*, 2009). Proteolysis in dry-cured ham occurs throughout processing, but at different rates and to varying extents depending on salt penetration and water migration. This difference implies greater proteolytic activity in the *biceps femoris* muscle compared to *semi membranosus* muscle, which will affect its texture (Parolari *et al.*, 1994; Virgili *et al.*, 1995; Rosell and Toldra, 1998; Virgili *et al.*, 1998). Proteolytic activity on dry-cured ham proteins is essentially attributed to cathepsins, which act for a longer time (Toldrà and Etherington, 1988; Toldrà and Flores, 1998).

However, in the first stage of processing, classical muscle ageing occurs, when calpains can also act. Skrlep *et al*. (2010) demonstrated the importance of pH of green ham on proteomic profile, lower pH favouring cathepsin activities. The time course of myofibrillar protein hydrolysis during the ripening process has been studied by one-dimensional gel electrophoresis (Toldrà *et al.,* 1997; Toldrà *et al.,* 1993; Monin *et al.,* 1997) and recently using protein labchip (Theron *et al.*, 2009) and by two-dimensional gel electrophoresis (Di Luccia *et al.,* 2005). Myosin heavy chains (MHC), myosin light chains (MLC1 and MLC2) and troponin C and I are targets of proteolysis. In the soluble fraction, the presence of tropomyosin shows that solubility properties have changed, possibly due to environmental conditions such as salt. In their study on the insoluble protein fraction of *semi membranosus* and *biceps femoris* muscles, Theron *et al*. (2011) noticed numerous of soluble proteins in the proteomic pattern of *semi membranosus*, indicating changes in solubility due to denaturing caused by the salt. We aimed to determine the proteomic pattern of insoluble protein fraction from *biceps femoris* muscle undergoing two salting conditions.

II – Material and methods

The study was based on a total of 60 pigs fed a cereal-based diet (60-80%), slaughtered at the Lahontan abattoir, and selected to meet the processing specifications of PGI Bayonne ham. The processing of Bayonne hams, which lasts 12 months in this experiment, was carried out at the Pyragena experimental station using the following sequence: salting, settling, oven drying, air drying, grease covering and ripening (Robert *et al.*, 2005). Sampling was carried out at 12 months, at the end of ripening. Proteomic analysis was performed on *biceps femoris*.

1. Insoluble protein extraction and electrophoresis

The method was adapted from Sayd *et al.* (2006). Frozen muscle was homogenized, using a glass bead agitator MM2 (Retsch, Haan, Germany), in 40 mM Tris HCl (pH 8) at 4ºC in a ratio of 1:8 (w/v). The homogenate was centrifuged at 4ºC for 10 min at 10,000 g. The supernatant was removed. The pellet was washed five times with this buffer to obtain only insoluble protein in low ionic strength buffer. After the last centrifugation, the supernatant was removed and the pellet was homogenized in 7 M urea, 2 M thiourea, 4% CHAPS (w/v), 1% DTT (w/v), at 4ºC in the same ratio as the first step. The homogenate was centrifuged at 4ºC for 10 min at 10,000 g. The supernatant, forming the insoluble protein fraction, was stored at –80ºC. The protein concentration was determined by the RC-DC assay (Bio-Rad). First 1 mg of proteins was incorporated in a buffer containing 7 M urea, 2 M thiourea, 2% CHAPS (w/v), 0.4% carrier ampholyte (v/v), 1% DTT (w/v), and bromophenol blue.

Samples were loaded onto immobilized pH gradient strips (pH 3-10 NL, 17 cm, Bio-Rad), and isoelectric focusing was performed using a Protean IEF cell system (Bio-Rad). Gels were passively rehydrated for 16 h. Rapid voltage ramping was subsequently applied to reach a total of 86 kVh. In the second dimension, proteins were resolved on 12% sodium dodecyl sulphate polyacrylamide gel electrophoresis (SDS-PAGE) gels using a Protean II XL system (Bio-Rad). Gels were stained with Coomassie Blue (colloidal blue). Three gels were produced per sample, giving 180 gels in all.

2. Image analysis and statistical treatment of data

Gels were visualized and analysed using the two-dimensional electrophoresis (2DE) image analysis software Samespots (non-linear dynamics). Aligned spots were normalized by expressing the relative quantity of each spot as the ratio of individual spot quantity to the total quantity of valid spots. Relative quantities were expressed in ppm. For one sample and one spot, the mean of three values (corresponding to the gels in triplicate) was calculated. The resulting set of average spot quantities underwent a one-way analysis of variance (ANOVA) using XLSTAT. A spot was considered significant when associated with $p < 0.05$ in ANOVA.

3. Protein identification by mass spectrometry

Was performed according to the method described by Theron *et al*. (2011). Coomassie stained spots of interest were manually excised using pipette tips. The spots were then destained, dehydrated and digested by trypsin. Peptide Mass Fingerprint (PMF) of trypsin digested spots were determined in positive-ion reflector mode using a Voyager DE Pro MALDI-TOF-MS (Applied Biosystems, Courtaboeuf, France). PMFs were compared to SwissProt (01/2008, 290 484 seq) protein sequence databases (ftp://ftp.ebi.ac.uk/pub/databases/uniprot/knowledgebase/uniprot_sprot.fasta.gz) using MASCOT 2.2 software [http://www.matrixscience.com]. When identification by MALDI-TOF proved unsuccessful, identification was also attempted using nano LC-ion trap MS/MS analysis. Identification of peptides was performed with Mascot 2.2, restricting the taxonomy to *mammalia* (20080417, 1177111 sequences) in the protein NCBInr database.

III – Results and discussion

Table 1 presented the protein identified by mass spectrometry. Proteins can be grouped according their biological function and some of presented different isoforms. 32 spots were significantly affected by salt; among them, 12 were entire proteins or protein fragments. Eight proteins or fragments belongs to the sarcoplamic proteins (creatine kinase, glycogen phosphorylase, peroxiredoxin, α crystalline, etc.).

Table 1: List of the spots identified by mass spectrometry (MALDI-TOF or LC/MS/MS)

Spot #	NCBI n°	Protein identity	Mascot score	%SC†	Matched peptides	Theoretical M/pI	Estimated M/pI	Conclusion
355	gi|790202	Skeletal alpha actin	113	38%	15	42024/5.23	37500/4.8	fragment
284	gi|790202	Skeletal alpha actin	85	33%	11	42024/5.23	37500/6.6	entire
285	gi|790202	Skeletal alpha actin	76	32%	12	42024/5.23	40000/6.0	entire
309	gi|790202	Skeletal alpha actin	91	24%	8	42024/5.23	40000/5.0	entire
258	gi|194018722	Muscle creatine kinase	162	45%	19	43032/6.61	42000/8.5	entire
283	gi|194018722	Muscle creatine kinase	146	44%	17	43032/6.61	39000/7.1	fragment
315	gi|194018722	Muscle creatine kinase	71	17%	6	43032/6.61	38000/6.8	fragment
322	gi|113205498	Enolase 3	79	39%	13	47100/8.05	38000/7.0	fragment
574	gi|106073338	Muscle glycogen phosphorylase	60	18%	14	83983/6.10	30000/7.0	fragment
695	gi|75063982	Alpha-crystallin B chain	65	38%	6	20116/6.76	20000/7.2	entire
510	gi|194044822	Similar to peroxiredoxin 4	88	46%	10	30536/6.01	30000/6.4	entire

†%SC corresponds to the percentage of sequence coverage.

Figure 1 presented the gel and the localisation of proteins associated to salt content. Some are involved in energy metabolism. Creatine kinase (spots 258, 283, 315), participates in energy transduction. Enolase is a glycolytic enzyme, responsible for the catalysis of the conversion of 2-phosphoglycerate (2-PG) to phosphoenolpyruvate (PEP), the penultimate step of glycolysis (spot 322). Glycogen phosphorylase (spots 574), is involved in carbohydrate metabolism. Peroxiredoxin is an antioxidant enzyme (spot 510) and α crystallin has chaperone-like properties including the ability to prevent the precipitation of denatured proteins. Two soluble proteins (α-crystalline and peroxiredoxine) presented higher quantity in low salt ham compared to the contralateral which had normal salt.

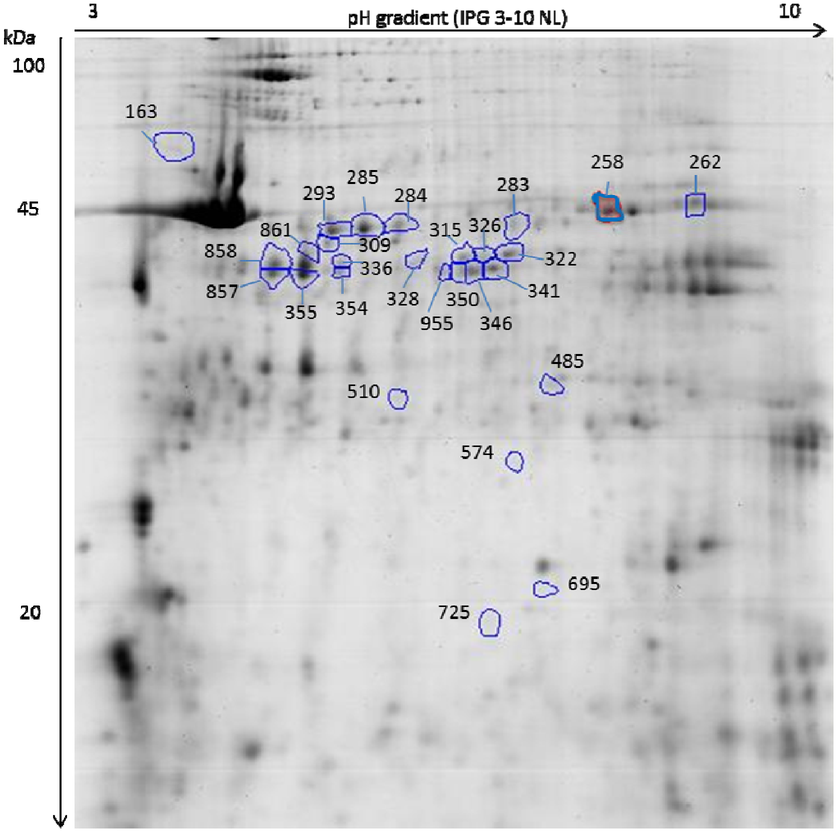

Fig. 1. Master gel and spots varying according to salt content.

The 4 remaining identified proteins were 3 isoforms of actin (spots 285, 284, 309) and one actin fragment (355). Numerous spots remained not identified. The actin spots revelead that the protein was not hydrolysed and exhibited higher quantity in normal salt ham. This could be due to a possible protection of the denaturated sarcoplasmic proteins of the complex actomyosin as it has been demonstrated in fresh meat. The insoluble fraction of protein revealed the presence of soluble proteins which may be explained by rapid denaturing due to salt, leading to precipitation onto the myofibrillar proteins and a jointed extraction. However In Iberian hams, Cordoba *et al*., (1994) reported that the loss of extractibility still occured during ripening. The dry cured ham having normal salt content (6.1%) showed higher entire actin and less antioxidant/chaperone proteins than the low salt content dry cured ham (5.3%). The fact that non hydrolysed proteins are still present after 12 months suggested that salt or co localization of soluble with myofibrillar proteins would bring a protection from the action of proteases.

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Physicochemical, hygienic and organoleptic characterisation of Slavonian Kulen, a traditional pork sausage from eastern Croatia

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Abstract. Slavonian Kulen (SK) is traditional dry sausage produced in the region of Slavonia in eastern Croatia. It is made from mince of pork, back fat, spices and salt filled into pork cecum. After stuffing, the sausages are cold-smoked and ripened-dried afterward for several months. Present work aimed to analyse some physicochemical and organoleptic traits of ripe SK as well as the safety of final product. The SK samples (n=12) from different small-scale manufactures were analysed. The following physicochemical attributes were recorded: moisture 38.2%±3.6, protein 35.0%±3.1, fat 23.7%±4.6, moisture/protein ratio 1.1±0.1, pH value 5.37±0.23 and water activity (aw) 0.82±0.02. Mean organoleptic scores, on five-point scale, were 3.7±0.6 for surface appearance, 3.4±0.6 for surface smell, 3.8±0.5 for consistency, 3.2±0.4 for inner smell, 3.0±0.7 for cross section quality, 3.3±0.5 for texture, 3.1±0.4 for taste and aroma, 3.0±0.5 for after taste and 3.2±0.4 for overall quality. Regarding the product safety, the following results (per kg) were determined: histamine 330.8 mg±126.3, tyramine 233.9 mg±124.7, nitrite 6.55 mg±3.88 and benzo(*a*)pyrene 0.05 µg±0.03. *Salmonella* spp. and *L. monocytogenes* have not been found in any sample while counts of *S. aureus*, enterobacteria and sulfite-reducing clostridia were in accordance with regulations.

Keywords. Pork – Dry sausages – Slavonian kulen – Physicochemical traits – Safety.

Caractérisation physico-chimique, hygiénique et organoleptique du Kulen slavonien, la saucisse de porc haché traditionnelle de Croatie orientale

Résumé. *Le Kulen slavonien (SK) est une saucisse sèche traditionnelle produite dans la région de Slavonie, située dans l'est de la Croatie. Elle est composée de maigre de porc, de graisse, d'épices, et de caecum de porc rempli de sel. Après remplissage, les saucisses sont fumées à froid, et ensuite laissées à sécher pendant quelques mois. Le travail présent visait essentiellement à analyser certaines caractéristiques physico-chimiques et organoleptiques de la production de SK, ainsi que la sécurité des produits finals. On a analysé les échantillons de SK (n=12) produits par différents fabricants en petite série. On a enregistré les attributs physico-chimiques suivants: humidité 38,2%±3,6, protéine 35,0%±3,1, graisse 23,7%±4,6, ratio humidité/protéine 1,1±0,1, valeur pH 5,37±0,23 et activité de l'eau (ae) 0,82±0,02. Les résultats organoleptiques moyens sur une échelle à cinq points sont les suivants: 3,7±0,6 pour l'apparition en surface, 3,4±0,6 pour l'odeur en surface, 3,8±0,5 pour la consistance, 3,2±0,4 pour l'odeur intérieure, 3,7±0,7 pour la qualité de la coupe transversale, 3,3±0,5 pour la texture, 3,1±0,4 pour la saveur et l'arôme, 3,0±0,5 pour l'arrière-goût et 3,2±0,4 pour la qualité générale. En ce qui concerne la sécurité des produits, on a déterminé les résultats suivants (par kg): histamine 330,8 mg±126,3, tyramine 233,9 mg±124,7, nitrite 6,55 mg±3,88 et benzopyrène 0,05 mg±0,03.* Salmonella *spp. et* L. monocytogenes *n'ont été constatés dans aucun échantillon, et par ailleurs les calculs concernant* S. aureus*, entérobactéries et bactérie clostridiale étaient en conformité avec les règlements.*

Mots-clés. *Porc – Saucisse sèche – Kulen slavonien – Caractéristiques physico-chimiques – Sécurité.*

I – Introduction

The Slavonian Kulen (SK) is a traditional pork sausage from Slavonia region in eastern Croatia that is produced seasonally at many households and small-scale manufactures. It is made from a mixture of selected and minced pork and back fat, salt and spices such as paprika and garlic filled into pork blind gut (cecum). After stuffing, the SK is cold smoked and ripened-dried afterward for several months until the shelf-stability and typical organoleptic properties are achieved. The quality of SK can be influenced by various factors, like pig breed, rearing and feeding conditions, pre-slaughter handling and post-slaughter conditions which all affect raw pork quality. Several other factors, like selection of lean meat and fat, addition of salt and spices, hygiene and environment (i.e. temperature, humidity, air velocity) during fermentation, smoking, drying and ripening may additionally contribute to diversity of product quality. As a result, the characteristics of final product including its safety may vary, as between producers, so between the years.

The aim of present work was to investigate some physicochemical and organoleptic attributes of traditional SK sausage, in order to better characterize it. In addition, some parameters of hygienic quality and safety of final product were assessed.

II – Materials and methods

Twelve ripe SK sausages aged about 6 months were sampled from different small-scale manufactures in Slavonia. All SK sausages were produced traditionally following the similar manufacturing steps and using the same type of ingredients as presented in scheme in Fig. 1.

After collecting samples were kept in cool until the analyses. The pH values were measured by TESTO 230 pH meter (TESTO®, Germany) by insertion of penetration electrode (type 13) in the core of halved sausage. Water activity (aw) was measured with the HygroPalm AW1 SET instrument (ROTRONIC®, Germany) using Aw Quick mode in samples, which were taken after coarse homogenisation of 80 g of the core of the sausage. The determination of moisture was done by heating the samples at 105°C until the constant weight. The nitrogen (N) content was determined by Dumas method and proximate protein content (%) was calculated using the conversion factor of 6.25 x N. Crude fat (%) was analysed by extraction using the Weibull-Stoldt method. Histamine and tyramine were analysed by thin-layer chromatography according to the procedure described by Macan *et al*. (2006). Nitrite content was determined according to HRN EN 12014-3 method. The microbial safety of product was assessed by the determination of the presence of *Salmonella* spp. and *Listeria monocytogenes* and the number of *Staphylococcus aureus*, enterobacteria and sulfite-reducing clostridia according to HRN EN ISO 6579, HRN EN ISO 11290-1, HRN EN ISO 6888-1, HRN ISO 21528-2 and HRN ISO 15213 method, respectively. Surface moulds were isolated according to the HRN ISO 7954 method. In addition, the contamination by polycyclic aromatic hydrocarbons (PAH) was assessed on sub-sample (n=5) by the determination of benzo(*a*)pyrene (B*a*P) using the thin-layer chromatography and spectrophotometer.

Organoleptic evaluation of samples was carried out by Faculty Department staff familiar with organoleptic evaluation of SK. Each assessor (n=4) was served with one freshly cut slice of SK (thick about 0.5 cm) on a white plastic plate for tasting, whereas the rest of the sausage half was exposed for visual inspection and touch. They were asked to evaluate on a scale from 1 (minimum grade) to 5 (maximum grade) surface appearance, surface smell, consistency, inner smell, cross section quality, texture, taste and aroma and aftertaste. Based on mean score of the particular organoleptic characteristic and the coefficient of importance for that characteristic, the overall quality was calculated by following formula:

Overall quality = $1/17 \times (a + b + c + 3d + e + 3f + 6g + h)$,

where a, b, c, d, e, f, g and h are the mean scores of evaluator's assessments for surface appearance, surface smell, consistency, inner smell, cross section quality, texture, taste and aroma, and after-taste, respectively. During the evaluation, the assessors were offered fresh water and apple slices freely.

For variables analysed the descriptive statistics (minimum, maximum, mean, SD - standard deviation, and CV - coefficient of variation) were calculated.

Manufacturing steps	Raw material and ingredients	Technological parameters
Slaughtering	Heavy pigs (>120 kg)	Carcass cooling, 24 h
Carcass cutting, meat and back fat selection	Leg (I category meat) Back (I category meat) Shoulder (II category meat) Neck (III category meat) Firm back fat	Lean met cleaned from fat and connective tissue
Cooling and draining of meat		Temperature around 5°C, 12-24 h
Mincing and mixing	← Ratio of I:II or II+III meat category = 80:20 ← Back fat ← Salt ← Hot red paprika ← Mild red paprika ← Garlic	Particle size 4-12 mm up to 10-20% 18 - 22 g/kg* up to 15 g/kg* up to 10 g/kg* up to 10 g/kg* *of meat and fat mixture
Stuffing and binding		Cleaned and washed pork cecum, binding with hemp rope
Fermentation and smoking		Natural fermentation, temperature 8 - 25 °C, 30 days
Drying and ripening		Temperature 10 -15 °C,150 - 180 days, weight loss 40 - 50%
Storage		Cool, dry and dark place

Fig. 1. Schema of traditional manufacturing processes of Slavonian kulen sausage.

III –Results and discussion

The results of organoleptic assessment, physicochemical and hygienic parameters, and microbiological analysis of SK are shown in Tables 1, 2 and 3, respectively.

In SK organoleptic evaluation (Table 1), the highest variability was found for cross section quality (CV=24.7%), while grades for the other traits varied similarly with quite uniform overall quality. The highest graded traits were consistency and surface appearance, and the lowest graded were cross-section quality and after-taste.

The CV of both *aw* and pH values (Table 2), which are commonly use in terms of shelf life and safety of dry sausages (Incze, 2007), was very low revealing the homogeneity of SK for these parameters. This is in agreement with result reported by Karolyi (2005). Generally, meat products are considered "shelf-stable" if have pH < 5.2 and *aw* < 0.95 or only pH < 5.0 or *aw* $<$ 0.91 (Leistner and Rödel, 1975). Hence, the ripe SK with an average *aw* value of 0.82 and pH value of 5.37 is shelf-stable product, principally due to extensive dehydration occurred.

European fermented sausages can be roughly classified as Mediterranean, or Southern Europe type, generally characterized by long ripening, slow pH fall with final pH > 5.0 and flavor significantly affected by the use of spices; and Northern Europe type, characterized by fast acidification, final pH < 5.0, smoking and short ripening (Zanardi *et al.*, 2004). Accordingly, SK is similar to long matured low acid Southern Europe type of fermented sausages with the addition of smoking. Similar pH and aw values to those observed for SK were reported for Majorcan sobrasada, which is also stuffed into pork cecum (Rosselló *et al*., 1995; Martínez *et al*., 2008).

Table 1. Descriptive statistics for organoleptic traits of Slavonian kulen

Trait	Min	Max	Mean	SD	CV (%)
Surface appearance	2.3	4.5	3.7	0.6	17.4
Surface smell	2.5	4.8	3.4	0.6	17.4
Consistency	3.0	4.8	3.8	0.5	13.0
Inner smell	2.5	3.8	3.2	0.4	12.4
Cross section quality	1.5	4.0	3.0	0.7	24.7
Texture	2.5	4.0	3.3	0.5	14.7
Taste and aroma	2.5	4.0	3.1	0.4	14.0
After taste	2.0	3.8	3.0	0.5	15.5
Overall quality	2.8	3.7	3.2	0.4	10.8

Table 2. Descriptive statistics for physicochemical and hygienic quality traits of Slavonian kulen

Trait	Min	Max	Mean	SD	CV (%)
pH value	5.07	5.75	5.37	0.23	4.3
aw†	0.79	0.85	0.82	0.02	2.2
Moisture (%, w/w)	31.7	42.8	38.2	3.6	9.4
Total fat (%, w/w)	16.4	31.0	23.7	4.6	19.3
Protein (%, w/w)	30.3	39.6	35.0	3.1	8.8
M/P††	1.0	1.3	1.1	0.1	9.4
Nitrite (mg/kg)	2.93	14.30	6.55	3.88	59.2
Histamine (mg/kg)	160.0	560.0	330.8	126.3	38.2
Tyramine (mg/kg)	67.0	400.0	233.9	124.7	53.3
BaP µg/kg†††	0.05	0.13	0.05	0.03	69.9

†Water activity ††Moisture/protein ratio; †††Benzo(*a*)pyrene.

The compositional parameters of SK were more variable (Table 2), especially in terms of fat content. This can be attributed to the differences in added fat and selection of more or less lean cuts by certain producers. High fat variability is also reported in other traditional sausages (Ambrosiadis *et al*., 2004). The moisture in SK (38.2 %) is higher than those cited by Salgado *et al*. (2006) for chorizo and other Spanish traditional sausages what could be explained by large diameter of SK. Compared to sausage of similar size and maturing time, e.g. Majorcan sobrasada (Rosselló *et al*., 1995; Martínez *et al*., 2008), the moisture in SK is also higher probably due to higher fat content in Majorcan sobrasada. In contrast, SK has less moisture than botillo sausage from Galicia region in northwest Spain (Lorenzo *et al*., 2000; García Fontán *et al*., 2007), which is also stuffed into cecum but it is ripened for much shorter time. Due to long drying and high lean meat share in the mince, the moisture and protein in ripe SK were similar (30-40 %) indicating high nutritional value of product. In fact, the average protein content in SK is higher than those usually found in other traditional sausages (Ambrosiadis *et al*., 2004; Salgado *et al*., 2006; Lorenzo *et al*., 2000; Moretti *et al*., 2004; Comi *et al*., 2005). Regarding to fat content reported for these products, the SK with 23.7 % is generally less fatty. The moisture to protein ratio (M/P), which indicates the extent of drying of lean meat part (Incze, 2007) was 1.1 (Table 2). The M/P, together with pH and aw, is used to distinguish semidry and dry sausages, i.e. an M/P of 1.2-1.3, which equals a*w* <0.89-0.90, is considered as a criterion for dry sausages (Incze, 2007). By that the SK could be clearly distinguished as dry sausage.

The SK is traditionally manufactured without nitrogenous salts. In meat curing, nitrite is widely used for antibacterial, colour and antioxidant purposes. On the other hand, high intake of nitrite presents a risk to human health due to its direct toxicity, or through the endogenous formation of carcinogenic nitrosamines (Sebranek, 2009). The average value of nitrite in ripe SK was 6.55 mg/kg what is lower than residual nitrite levels reported in sausages fermented with the use of nitrite/nitrate (Comi *et al*., 2005). High level of biogenic amines in foods is another issue of public health concern because of their potentially toxicological effects (Vidal-Carou *et al*., 2007). Biogenic amines are mainly produced by microbial decarboxylation of amino acids (Silla Santos, 1996) and could be found in various fermented and seasoned foods. The fermentation of sausages in particular offers optimal conditions for biogenic amine accumulation due to availability of free amino acids, the presence of micro-organisms and acidic environment that favours their aminogenic activity (Bover-Cid *et al*., 1999). The most frequent and most abundant biogenic amine usually found in fermented sausages is tyramine with average concentrations from 100 to 200 mg/kg (Vidal-Carou *et al*., 2009). Large diameter and long ripening of the sausage can contribute to a greater accumulation of tyramin (Bover-Cid *et al*., 1999; Parente *et al*., 2001; Miguélez-Arrizado *et al*., 2006; Komprda *et al*., 2009). This could explain the generally high levels of tyramine (Table 2) observed in SK (≥200 mg/kg in 75% of samples). The same factors may also be responsible, at least partly for the high accumulation of histamine observed (≥200 mg/kg in more than 90 % of samples). However, histamine is rarely found in fermented sausages manufactured under proper hygienic conditions and the occurrence of excessive levels of this biogenic amine is rather an indicator of defective hygienic conditions of raw materials and/or manufacturing processes (Vidal-Carou *et al*., 2007; Vidal-Carou *et al*., 2009).

Smoking of SK is done by thermal combustion of hardwoods, e.g. beech, ash, hornbeam. Logs and sawdust are sometimes used wet to lower the temperature of smoke. Smoking is usually in the chamber where the smoke is generated. Direct smoking, however, can lead to greater deposition of potentially unhealthy substances from the smoke, like polycyclic aromatic hydrocarbons (PAH) on the product surface (Andrés *et al*., 2007). Some PAH, like benzo(*a*)pyrene (B*a*P) are classified as possible human carcinogens (US EPA, 2002). The B*a*P is used as an indicator of total PAH presence in smoked foods and in EU the maximum B*a*P level of 5 μg/kg has been established for smoked meats (European Commission, 2005). In present work, the maximum B*a*P level in SK was far below this margin (Table 2) and lower than those reported for other traditional (Lorenzo *et al*., 2010) and industrial sausages (Djinovic *et al*., 2008). Possible explanations for low PAH contamination of SK could be the low temperatures of combustion which generate smoke with less PAH (Šimko, 2009), no use of softwoods which are high in resin that increases the PAH concentration in smoke (Stumpe-Viksna *et al*., 2008), and the low surface/mass ratio of SK which less favour the PAH adsorption (Lorenzo *et al*., 2010).

During fermentation and ripening of sausages various microorganisms which are not involved in fermentation are usually progressively eliminated by acidification and drying. As a result, dry fermented sausages are generally safe products which have rarely been involved in food contamination outbreaks. This is particularly true for industry where high sanitary standards and starter cultures are used to control the production. On the other hand, the traditional production is often connected with large variability in raw materials, operation units, fermentation, and/or ripening conditions and hygiene which may result in increased ability of pathogens to survive in end product (Skandamis and Nychas, 2007). For example, investigating the traditional production of sausages in households in Croatia, Kozačinski *et al*. (2008) found the increased total bacterial count at surfaces and equipment used in the sausage preparation including the finding of *S. aureus* and *Enterococcus faecalis* in some cases. The same pathogens, together with enterobacteria and sulfite-reducing clostridia were isolated in high counts from several samples of raw sausages for cooking, and some of them like *S. aureus* and sulfite-reducing clostridia were found to be exceedingly high in few samples of dry sausages. Close relationship between microbial ecosystems of traditional processing plants (so called "house flora") and produced sausages was also established in studies in other countries (e.g. Lebert *et al*., 2007).

Table 3. Results of microbiological analysis of Slavonian kulen

Sample	SRC cfu/g	S cfu/25g	E cfu/g	LM cfu/25g	SA cfu/g	Surface moulds
1.	< 10	neg.	< 10	neg.	< 10	neg.
2.	< 10	neg.	< 10	neg.	< 10	*Aspergillus flavus*
3.	< 10	neg.	< 10	neg.	< 10	neg.
4.	< 10	neg.	< 10	neg.	< 10	*Penicillium spp*.
5.	< 10	neg.	< 10	neg.	< 10	neg.
6.	< 10	neg.	< 10	neg.	< 10	neg.
7.	< 10	neg.	< 10	neg.	< 10	*Aspergillus glaucus*
8.	< 10	neg.	< 10	neg.	< 10	neg.
9.	< 10	neg.	< 10	neg.	< 10	*Penicillium spp.*
10.	< 10	neg.	< 10	neg.	< 10	neg.
11.	< 10	neg.	< 10	neg.	< 10	neg.
12.	< 10	neg.	< 10	neg.	< 10	neg.

SRC - Sulphite-reducing clostridia; S - *Salmonella spp*.; E – *Enterobacteriaceae*; LM – *Listeria monocytogenes*; SA – *Staphylococcus aureus*.

In present study (Table 3), the counts of *S. aureus*, enterobacteria and sulfite-reducing clostridia in all samples were in accordance with Croatian regulations (Narodne novine, 2004), while *Salmonella* spp. and *L. monocytogenes* were not isolated from any sample. Some of the mentioned meat contaminants, like enterobacteria are known for their high ability for biogenic amine production (Vidal-Carou *et al*., 2007). Hence, the high accumulation of histamine observed in this study may indicate the bacterial contamination of raw materials or hygienic failure during the early steps of sausage production, regardless of the absence or low levels of aminogenic microorganisms in final product. Similar to other fermented products which go through the long ripening, the fungal colonization of surface may also appears in SK.

The natural moulding, which results from contamination by environment-contaminating species, mainly from genera *Penicillium* and *Aspergillus*, may be desirable as it protects against the excessive drying and lipid oxidation and contributes to the flavour development (Spotti and Berni, 2007). On the other hand, many moulds have ability to produce mycotoxins and some of species that were isolated from SK (Table 3), i.e. *Aspergillus flavus*, do have toxigenic potentiality (Bailly and Guerre, 2009).

The presence of certain fungi is not always followed by toxin production as conditions (a*w* in particular) which allow toxin production are more restricted than those which give way to growth (Northolt *et al*., 1996). However, the presence of mycotoxins in both surface and deeper layers of naturally moulded SK has been recently reported by Frece *et al*. (2010). The contamination of spices and additives used in meat processing may additionally represent a source of mycotoxins (Bailly and Guerre, 2009).

IV – Conclusions

By the manufacturing steps and final characteristics, the SK is similar to long matured Southern Europe type of fermented sausages with the addition of smoking. According to final pH, a*w* and M/P, the SK could be characterised as low-acid dry sausages which shelf-stability is primarily conditioned by low a*w* of final product. The organoleptic traits were within the limits of characteristics specific to traditional production. Compared to similar products, the SK has higher protein content and less fat. All samples were microbiologically valid, with low nitrite level and PAH contamination but with high biogenic amines content and, in some cases, with the presence of potentially toxigenic moulds. Hence, from the hygienic and technological point of view, efforts should be made to improve sanitary standards and good manufacturing practice among producers. In addition, better characterization and control of typical microflora during the SK processing is essential in terms of its safety, acceptability and organoleptic quality.

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Effect of the slaughterhouse on behaviour, blood parameters, meat quality and raw ham defects in heavy pigs

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Abstract. The effect of different pre-slaughter procedures on behaviour, blood parameters, meat quality traits and raw ham defects were evaluated on 120 pigs reared in one farm and delivered in groups of 40 subjects to three slaughterhouses. Due to the different attitude of the personnel involved, differences in handling were evident at loading and at unloading where the difficulties to drive the pigs increased the behavioural events. Blood analysis parameters showed that different resting time did not reduce the physical stress experienced by the pigs, which seems related "per se" to loading, journey condition and unloading and not to the different handling procedures applied in each slaughter plant. Among the meat quality traits measured, the rate and the extent of post mortem pH decline and L* and a* colour coordinates were affected by the slaughterhouse. The presence and the seriousness of the veining defect was significatively affected by the slaughter condition while the "red skin" defect did not varied accordingly to the plant. The results of the present study showed that the "slaughter plant" effect, namely how the animals are handled before death, has an unquestionable responsibility to the final quality traits of both meat and raw ham.

Keywords. Pig – Behaviour – Blood parameters – Meat quality – Raw ham quality.

Effet de l'abattoir sur le comportement, les paramètres sanguins, la qualité de la viande et les défauts du jambon de porcs lourds

Résumé. *L'effet des procédures de pré-abattage sur le comportement, les paramètres sanguins, la qualité de la viande et les défauts du jambon, a été évalué sur 120 porcs élevés dans une ferme et livrés en groupes de 40 sujets à trois abattoirs. En raison de l'attitude différente du personnel concerné, les différences de traitement étaient évidentes au chargement et au déchargement où les difficultés à conduire les porcs ont augmenté les événements de comportement. L'analyse des paramètres sanguins a montré que les différents temps de repos n'ont pas réduit le stress physique subi par les porcs, qui semble lié "en soi" aux conditions de chargement, voyage, et déchargement, et non aux différentes procédures de manipulation appliquées dans chaque abattoir. En ce qui concerne* les *caractéristiques de qualité de la viande, seules la vitesse et l'ampleur de l'abaissement du pH post mortem ont été influencées par l'abattoir. La présence et la gravité du défaut de la veine ont été significativement influencées par l'abattage, tandis que le défaut de la "peau (couenne) rouge" n'a pas varié en fonction des différents abattoirs. Les résultats de la présente étude ont montré que l'effet de l'abattoir, c'est-à-dire la façon dont les animaux sont traités avant l'abattage, a une responsabilité indiscutable pour la qualité finale de la viande et du jambon frais.*

Mots-clés. *Porc – Comportement – Paramètres sanguins – Qualité de la viande – Qualité du jambon cru.*

I – Introduction

Impaired pig carcass, meat and raw ham quality after slaughter are often attributed to differences in responses to pre-slaughter procedures related to the "farm of origin" effect, which accounts for genetic type, feeding and housing conditions. This generic attribution leads to a difficult assignment of responsibility for inadequate pre-slaughter handling, which usually flows from the slaughterhouse to the truck driver and finally reaches the farmer. In order to better define the role played by the pre-slaughter treatments related to "farm of origin", the present study was carried out on heavy pigs reared on one farm and slaughtered in three different plants.

II – Materials and methods

Behaviour, blood parameters and quality traits of meat and raw hams of 120 pigs (LW kg 160 ± 9) of the same farm and subjected to different pre-slaughter handling were examined. Pigs were delivered the same day in three groups of 40 subjects and slaughtered in three different plants (A, B and C). During each delivery, the time of loading, transport, unloading, and resting were recorded (Table 1).

Table 1. Number of pigs (castrated males, females) and time (min) of pre-slaughter procedures in the three plants

Plant	No. Pigs	Loading	Transport	Unloading	Resting	Total
A	40 (18,22)	32	50	7	81	170
B	40 (19,21)	17	101	5	15	138
C	40 (17,23)	15	68	9	60	152

The transport was carried out by three similar decks vehicles (space allowances 0.50 m² - 0.55 m² per 100 kg of LW). At the end of the journey, each group was unloaded and droved to the resting pens where watering nipples were available. Use of electric prods and mixing between subjects of different pens never occurred. During loading and unloading, the number of reversals, balks, falls, slips, bites, jumps, evacuations and vocalizations were recorded for each group. In the resting pens, one observation of 1 min each 15 min was carried out to record postures (standing, sitting, lying down), exploratory, olfactory and aggressive actions as well as drinking, mastication and evacuation. According to the resting time, the observations were 5 at the plant A, 1 at the B and 4 at the C.

In the slaughterhouse B the pigs were showered during resting with cold water for 6 min. The pigs were electrical stunned by manual tongs in the A and C plants (V 180-220) and by automatic system (V 300) in the B plant. At sticking, the blood was collected from each pig, centrifuged at 3500 g for 15 min and frozen at -20°C until analysis. Albumin, lactate, glucose, total protein, urea, osmolarity and creatin kinase contents were determined by an automatic analyzer (BM HITACHI 911 - Roche) and cortisol by enzyme-immunoassay with the automatic analyser DPC Immulite (Medical System) and the COR LKCO1 kit (Medical System). The weight of hot carcasses and left thighs after trimming were recorded. The pH_1 (1 h *post mortem*) and pH_u (24 h *post mortem*) measures as well as colour coordinates (L*,a*,b*) were recorded on *semimembranosus* and *bices femoris* muscles. On all left thighs, an expert assessor carried out the scoring of veining (Russo *et al*., 2003) and "red skin" (Lo Fiego *et al*., 2006) defects (Table 5). Based on these classifications, the average scores and percentage of thighs in each class were calculated.

The distribution of behavioural events and frequencies of postures were compared by χ^2 (Fischer's exact test). Creatin kinase, cortisol, lactate and osmolarity data were normalised by a log10 transformation and glucose by inverse transformation. Plasma parameters, carcass and meat characteristics, were processed by a model including the fixed effects of slaughterhouse, sex and their interaction, the latter excluded when P>0.05. Differences due to slaughterhouse and sex regarding the distributions of the thighs among the scoring classes were examined by non- parametric test of Wilcoxon for the scores and by χ^2 (Fisher exact test).

III – Results and discussion

A significant difference (P<0.05) in the distribution of behavioural events between pigs addressed to plants A and C was found (Table 2).

Table 2. Behavioural events observed at loading and at unloading in the three plants

Events	Loading			Unloading		
	Plant A	Plant B	Plant C	Plant A	Plant B	Plant C
	N	N	N	N	N	N
Reversal	4	9	21	1	2	7
Balk	3	16	20	-	2	-
Fall	1	1	1	-	2	2
Slip	4	5	5	-	6	-
Vocalization	-	10	15	-	10	18
Total	12	41	62	1	22	27
No. pipe/pig	1.6	4.1	4.8	0.2	0.7	4.8

A progressive raise of events was observed accordingly with the reduction of the loading time. They increased from 12 for the pigs destined to the plant A to 62 for those addressed to the plant C. The raise of behavioural events regarded mainly reversals and vocalizations. The difficulties to move the pigs towards the vehicle increased the use of the rubber pipe that, in turn, leads to more vocalizations. The observations recorded at loading highlight that differences in pre-slaughter handling raises very soon and confirms that fast driving increases the difficult to handle the pigs (Grandin, 2007). Despite very similar facilities and duration of unloading, there were significant differences (P<0.01) between the plants B and C in the distribution of the behavioural events. The higher number of events recorded in the latter plant, particularly the vocalization, could be related to a greater stimulation of movement, as showed by the higher use of the rubber pipe. In general, these results show that at loading and at unloading the differences in behaviour mainly reflected different personnel attitude to carry out such operations. The frequency of the postures (data not showed) was similar in all slaughter plants, even if there is a significant difference (P<0.05) in the first observation between B and C plants, the latter characterized by more sitting pigs. The behavioural events observed in the active subjects were limited to the exploration only. At the second observation, the frequencies of posture were not statistically different (P>0.05) between A and C plants, while, to respect to the previous one, an increase of subjects lied down was observed in both plants. Moreover, all the active subjects manifested exploratory activities only, confirming what was recorded in the first one. The subsequent observations found all pigs lied down until the stunning.

Least squares mean values of plasma variables are shown in Table 3. They were within the normal range (Boyd, 1984) in all groups excepted for the higher lactate and creatin kinase values. The levels of albumin, osmolality and total protein were higher (P<0.05) in the plasma of pigs slaughtered in plants A and C vs those slaughtered in plant B. This might indicate a week dehydration (Knowles and Warriss, 2007) in the former pigs, probably related to the longer resting time and to the absence of stimuli to drink, as the behavioural observation shown. In addition, the lower glucose and the higher lactate levels observed in pigs slaughtered in A and C plants (P<0.05) could be due to the longer lairage, even if their physical activity was limited to the exploration. CK activity was found very high in all groups. Irrespectively of the differences in pre-slaughter procedures, the means values did not differ significantly among the groups.

Table 3. Effect of slaughterhouse on plasma variables (untransformed least squared means ± SE)

Trait		Plant A	Plant B	Plant C
Albumin	g/l	42.90^{b} ± 0.54	39.93^{a} ± 0.54	45.33^{c} ± 0.54
Lactate	mmol/l	16.08^{a} ± 0.72	9.47^{b} ± 0.71	12.19^{c} ± 0.73
Glucose	mmol/l	6.22^{ab} ± 0.23	7.00^{b} ± 0.23	5.46^{a} ± 0.24
Total protein	g/l	79.51^{b} ± 0.76	69.50^{a} ± 0.76	76.70^{c} ± 0.79
Cortisol	nmol/l	231.29 ± 0.03	201.89 ± 0.03	233.70 ± 0.03
Urea	mmol/l	5.34 ± 0.16	5.26 ± 0.16	5.29 ± 0.17
Osmolarity	mOsm/K	309.18^{b} ± 1.19	300.48^{a} ± 1.17	310.35^{b} ± 1.22
Creatin kinase	U/l	1536.85 ± 140	1610.53 ± 138	1525.97 ± 144

a, b, c: P<0.05.

This result suggests that in terms of reaction to the physical stress experienced before lairage, the examined pigs showed a similar response. The prolonged resting time in A and C plants was not sufficient to recover from this stress.

The groups of pigs delivered to the three slaughterhouses were similar for carcass and trimmed thigh weight (Tables 4 and 5).

Table 4. Effect of slaughterhouse on carcass weight and meat quality traits (means ± SE)

	Plant A	Plant B	Plant C
Carcass weight (kg)	130.5 ± 1.14	132.2 ± 1.13	131.8 ± 1.15
M. semimembranosus:			
pH_1	6.12^{a} ± 0.04	6.53^{b} ± 0.04	6.09^{a} ± 0.04
pH_u	5.42^{a} ± 0.01	5.53^{b} ± 0.01	5.52^{b} ± 0.01
L*	47.99^{b} ± 0.45	47.83^{b} ± 0.46	45.36^{a} ± 0.47
a*	11.64^{c} ± 0.25	9.15^{a} ± 0.25	10.20^{ab} ± 0.25
b*	0.23 ± 0.31	0.22 ± 0.31	0.40 ± 0.31
M. biceps femoris			
pH_u	5.45^{a} ± 0.01	5.54^{b} ± 0.01	5.50^{ab} ± 0.01
L*	47.6^{b} 0 ± 0.34	46.92^{b} ± 0.35	43.46^{a} ± 0.35
a*	13.01^{b} ± 0.26	11.62^{a} ± 0.27	12.70^{ab} ± 0.27
b*	1.84	1.65	1.90

a, b, c: P<0.05.

Sex showed a significant effect on carcass weight, which was higher (P<0.01) in castrated males (data not shown), while there was no significant interaction between sex and slaughter. The slaughterhouse significatively affected the rate and the extent of *post mortem* acidification and the colour coordinates except for b* values. Nevertheless, the differences among plants were little and there were not defects or tendency toward PSE and DFD defects.

Regarding the veining defect (Table 5), the slaughterhouse showed a significant effect (P <0.01) on both average score and distribution of thighs among the scoring classes. The highest frequency and seriousness of the veining defect were found in plant B where, although with different intensity, 100 % of the trimmed thighs showed this defect. The score of the "red skin" defect was not significantly affected by the slaughter plant, nor was the distribution of thighs among the three scoring classes.

Table 5. Effect of slaughterhouse on distribution (%) of the left thighs among the classes of veining and "skin red" defects within slaughterhouses (means ± S.E.)

	Plant A	Plant B	Plant C	Signif.
Trimmed thigh weight (kg)	13.8 ± 0.14	13.9 ± 0.14	14.3 ± 0.15	ns
Veining score	2.15	2.77	2.54	**
"Red skin" score	2.27	2.40	2.10	ns
Veining score classes:				**
- 1 (no defect) (%)	20.0	0.0	2.5	
- 2 (light) (%)	47.5	30.0	40.0	
- 3 (evident) (%)	30.0	62.5	57.5	
- 4 (serious) (%)	2.5	7.5	0.0	
"Red skin" score classes:				ns
- 1 (absent) (%)	7.5	2.5	10.0	
- 2 (light) (%)	57.5	55.0	70.0	
- 3 (severe) (%)	35.0	42.5	20.0	

ns: not significant; **: P<0.01

However, the negative effect exerted by the slaughter conditions at plant B was confirmed, where the incidence of thighs with severe "red skin" reached 42.5 %, while in plants A and C this reached 35.0 % and 20.0 % respectively. Sex did not show any significant effect on the two defects. The results of the present study showed that the slaughter plant, namely how the animals are handled before and after death, plays an important role on the incidence and severity of the veining defect. This confirms previous findings on the role of some treatments *pre-* and *post mortem* such as the lairage duration, the method of stunning and the pre-chilling time of the thighs (Lo Fiego *et al*., 2003; Nanni Costa *et al*., 2005).

IV – Conclusions

The results achieved show that, irrespectively of the common origin of examined pigs, differences in pre-slaughter handling and, consequently, differences in pre-slaughter behaviour raise very soon, mainly related to the attitude to the personnel in animal driving. The most evident consequences concern the behaviour of animals and the metabolic responses to the resting, while the physical stress related to the pre-slaughter handling seems less affected by different pre-slaughter procedures. The experimental plan used for the present study did not allow identification of specific factors or treatments relating to the slaughter that could be responsible for the differences observed among plants. However, the results give an unquestionable responsibility to the slaughterhouse for the presence and seriousness of the veining defect. On the contrary, the incidence and severity of the "red skin" defect was not significantly related to the slaughter. This result suggests the need to investigate factors in the farm pens that may affect the condition of the thigh skin.

Acknowledgments

This work was supported by the Emilia-Romagna Region (Italy).

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Effect of the use of chestnuts in the finishing diet on fatty acid profile in different tissues of the Celta pig breed

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Abstract. The effect of the use of chestnuts in the finishing diet on the fatty acid profiles of different tissues (muscles and liver) of the Celta pig (an autochthonous breed from the NW of Spain) was studied. Thirty six pigs were separated in three different groups according to the type of feeding during the finish-fattening period (three months): commercial compound feed, mixture of the chestnuts and commercial compound feed, and chestnuts. Fatty acid composition of neutral (NL) and polar lipids (PL) in the intramuscular fat of *Longissimus dorsi* (LD) and *Psoas maior* and *minor* (PMM) muscles and in the hepatic fat were analysed. The predominantly oxidative PMM muscle had higher polyunsaturated and lower monounsaturated fatty acids contents than LD both in NL and PL. The oleic acid (C18:1 n-9) content in neutral lipids and linoleic (C18:2 n-6) and linolenic (C18:3 n-3) acid contents in polar lipids showed significant higher values in the LD and PMM muscles from pigs fed only with chestnuts. Also, the inclusion of chestnuts in the diet significantly affected the palmitic (C16:0), stearic (C18:0) and oleic (C18:1 n-9) acid contents in both lipid classes in liver; the arachidonic (C20:4 n-6) and linolenic (C18:3 n-3) acids contents were also affected in NL and PL, respectively.

Keywords. Celta pig breed – Lipids – Finishing diet – Fatty acid – Chestnuts.

Effet de l'utilisation de la châtaigne dans l'alimentation de finition sur le profil en acides gras dans les différents tissus du porc de race Celta

Résumé. *L'effet de l'utilisation de la châtaigne dans l'alimentation de finition sur les profils en acides gras de différents tissus (muscles et foie) de porcs Celta (une race autochtone du nord-ouest de l'Espagne) a été étudié. Trente-six porcs ont été séparés en trois groupes différents selon le type d'alimentation au cours de la période de finition-engraissement (trois mois): aliment composé commercial, mélange de châtaignes et d'aliment composé commercial, et châtaignes. La composition en acides gras des lipides neutres (LN) et polaires (LP) dans la graisse intramusculaire des muscles* Longissimus dorsi *(LD) et* Psoas maior *et* minor *(PMM) et dans la graisse hépatique a été analysée. Le muscle PMM, avec un métabolisme principalement oxydatif, a montré des teneurs plus élevées en acides gras polyinsaturés et plus basses en acides gras monoinsaturés que le muscle LD, tant pour les LN que pour les LP. La teneur en acide oléique (C18:1 n-9) dans les lipides neutres et la teneur en acides linoléique (C18:2 n-6) et linolénique (C18:3 n-3) dans les lipides polaires montrent des valeurs plus élevées, avec une différence significative, dans les muscles LD et PMM des porcs nourris seulement avec des châtaignes. En outre, l'inclusion des châtaignes dans l'alimentation a affecté d'une façon significative les teneurs en acides palmitique (C16:0), stéarique (C18:0) et oléique (C18:1 n-9) dans les deux classes de lipides dans le foie; les teneurs en acides arachidonique (C20:4 n-6) et linolénique (C18 3 n-3) ont également été affectées dans les LN et LP, respectivement.*

Mots-clés. *Race porcine Celta – Lipides – Alimentation de finition – Acides gras – Châtaignes.*

I – Introduction

Celta pig is an autochthonous swine breed from the NW of Spain, characterized by its rusticity and its adaptation to the environment. It is catalogued as Special Protection pig breed in danger of extinction (BOE 21/11/1997, R.D. 1682/1997) because was substituted with the arrival of the commercial crossbreds with higher productive capacity during the 1990s. At the present time, this breed is recovering and the carcasses are used in the production of dry meat products which have a high value on the market.

The muscles differ in the amount and fatty acid composition of the main lipid fractions, neutral lipids and phospholipids. The influence of diet on the fatty acid composition of animal tissues, particularly muscle and adipose tissue, has been the subject of much investigation (Wood *et al*., 2008). An efficient way of influencing the fatty acid composition in pork is by feeding sources with varying fatty acid composition. The desired fatty acid composition in meat products should give appropriate pork quality: shelf life, flavour and high nutritional value (Hallenstvedt *et al*., 2010).

The use of the chestnuts (NW region is the main area of production in Spain) in the feeding of the Celta pig breed, in a extensive management system, would allow reducing the production costs and puting in the market quality products, differentiated, with a high added value and with healthier fat. In previous studies (Franco *et al*., 2006; Martínez *et al*., 2007) were investigated the fatty acid profile of the total, neutral and polar lipids in different deposits of Celta pigs fed with traditional diet.

The objetive of this study was to assess the effect of the inclusion of chestnuts in the finishing diet on the fatty acid composition of neutral and polar lipids in intramuscular and hepatic fat of Celta pigs.

II – Materials and methods

1. Pigs, samples and diets

In order to carry out this study, 36 castrated Celta pigs (males and females) were fed in three different groups: A) Fed during all their life (16 months) with commercial compound feed, B) Fed with commercial compound feed the first 12 months and with a mixed (commercial compound feed/chestnuts) diet in the last four months before slaughtering and C) Fed with commercial compound feed the first 12 months, with a mixed (commercial compound feed/chestnuts) diet the 13th month, and receiving only a chestnut diet in the last three months before slaughtering. After slaughtering, and after 24 hours of refrigeration, were obtained in each carcass samples from intramuscular fat – *Longissimus dorsi* (LD) and *Psoas maior* and *minor* (PMM) muscles – and hepatic fat.

Chestnuts and commercial compound feed were sampled and was determined the chemical composition according to the Association of Official Analytical Chemist (1990) (Table 1). The fatty acids profile was determined as shown in the following paragraph.

2. Analytical methods

The fat of the samples was extracted following the procedure described by Folch *et al*. (1957). The neutral and polar lipids from muscles and liver samples were obtained according to the procedure developed by Kaluzny *et al*. (1985). Fat extracts were methylated and fatty acid profiles of the both lipids were determined using the procedure described by Franco *et al*. (2006). Fatty acid methyl esters were analysed by Gas Chromatography using a Thermo Finnigan Trace GC (Thermo Finnigan Trace GC (Thermo Finnigan, Austin, TX, USA) The separation of the different fatty acids was carried out in an Innowax column: 30 m; 25mm ID; 0.25 mm film thickness (Agilent Technologies, Palo Alto, CA, USA). The temperature of the detector was 250ºC and that of the injector 230ºC. The gasses used were air (350 mL/min), hydrogen (335 mL/min) and helium (carrier gas) (30 mL/min). Results are expressed as percentages of the total fatty acid composition. All analyses were carried out in duplicate.

Table 1. Chemical composition (expressed as g/100 g) and fatty acids of chestnuts and commercial compound feed

	Chestnut	Compound feed
Dry matter	51.9	89.5
Crude protein	4.2	15.3
Eter extract	3.3	4.9
Crude fiber	2	4.6
Starch	32	39.7
Fatty acids (g/100 g total fatty acids)		
C12:0	0.03±0.00	0.16±0.01
C14:0	0.15±0.00	1.28±0.06
C14:1	N.D.	0.12±0.01
C15:0	0.10±0.00	0.16±0.01
C15:1	N.D.	0.04±0.00
C16:0	15.88±0.04	20.83±0.63
C16:1	0.48±0.00	1.75±0.06
C17:0	0.13±0.00	0.42±0.00
C17:1	0.06±0.00	0.21±0.00
C18:0	1.41±0.02	9.00±0.32
C18:1 *n-9*	26.05±0.07	24.85±0.30
C18:2*n*-6	40.47±0.32	24.04±0.00
C18:3*n*-3	5.94±0.09	2.58±0.04
C20:0	0.28±0.00	0.15±0.00
C18:3*n*-6	0.41±0.00	0.41±0.01
C20:2*n*-6	0.22±0.02	0.03±0.00
C20:3*n*-6	0.17±0.06	0.01±0.00
C20:4*n*-6	2.57±0.02	7.03±0.11
C24:0	5.65±0.14	6.93±0.01

3. Statistical analysis

Data was subjected to a two-way analysis of variance (ANOVA) using the General Linear Model procedure of the computer programme Statistica© 5.1 for Windows (Statsoft Inc., Tulsa, OK, USA) to determine the overall effect of muscle (LD *vs* PMM) and diet (commercial compound feed, mixture of the chestnuts and commercial compound feed, and chestnuts).

III – Results and discussion

Fatty acid compositions of NL and PL from LD and PMM muscles are shown in Table 1. In NL, LD muscle showed higher levels ($P<0.05$) of UFA and MUFA and lower ($P<0.05$) of PUFA than PMM. These differences are a direct consequence of higher amounts of palmitoleic (C16:1) and oleic (C18:1 n-9) acids in LD muscle, and higher amounts of palmitic (C16:0), linoleic (C18:2 n-3), C20:2 n-6, arachidonic (C20:4 n-6) and C20:3 n-6 acids in PMM muscle.

The total amount of MUFA and PUFA in PL was also affected by the type of muscle, PMM muscle showing higher levels ($P<0.05$) of PUFA and lower ($P<0.05$) of MUFA than in the NL. These differences are a direct result of higher proportions of linoleic (C18:2 n-3), linolenic

(C18:3 n-3), arachidonic (C20:4 n-6) and C20:3 n-6 acids in PMM muscle and higher proportions of oleic (C18:1 n-9) acid in LD muscle.

PMM is a predominantly oxidative muscle, showing in both lipids class higher levels of PUFA than LD, which has been described as a predominantly glycolytic muscle in the scientific literature (Muriel *et al*., 2002). With regards to other publications studying the effect of the metabolic type muscle on the fatty acid composition of NL and PL is some controversy (Andrés *et al*., 2001; Hernández *et al*., 1998; Alasnier *et al*., 1996; Leseigneur-Meynier and Gandemer, 1991). Leseigneur-Meynier and Gandemer (1991) have found a similar tendency: oxidative muscles have higher PUFA contents in PL and NL.

The fatty acid composition of NL and PL of liver are shown in Table 2. In NL, liver had a higher percentage of PUFA than LD and PMM muscles. These differences are a direct consequence of higher amounts of linoleic (C18:2 n-3) and arachidonic (C20:4 n-6) acids in liver. The profile of fatty acid that we observed in the liver coincides with that found in a previous study by Martínez *et al*. (2007).

Table 2. Fatty acid composition, % and standard error of the mean (SEM), of neutral and polar lipids in LD and PMM muscles

	Neutral lipids (NL)				**Polar lipids (PL)**			
	LD		***PMM***		***LD***		***PMM***	
	Mean	**SEM**	**Mean**	**SEM**	**Mean**	**SEM.**	**Mean**	**SEM**
C12:0	0.05	0.006	0.05**	0.006	0.02$_a$	0.005	0.01$_b$	0.002
C14:0	1.37	0.016	1.39	0.021	2.45$_a$	0.138	1.87$_b$	0.149
C14:1	0.03$_a$	0.002	0.04$_b$	0.003	1.03	0.174	0.71	0.136
C16:0	24.97$_a$	0.139	26.03$_b$	0.160	28.20$_a$	0.279	26.29$_b$	0.329
C16:1	3.62$_a$	0.091	3.01*$_b$	0.089	1.16$_a$	0.038	0.84$_b$	0.035
C17:0	0.20$_a$	0.008	0.31$_b$	0.013	0.23*	0.014	0.25	0.015
C17:1	0.19$_a$	0.006	0.22$_b$	0.009	1.16	0.113	1.31	0.142
C18:0	13.37*$_a$	0.289	14.35$_b$	0.256	5.13**$_a$	0.185	6.93$_b$	0.289
C18:1n-9	46.21*$_a$	0.407	40.76**$_b$	0.423	14.30$_a$	0.366	10.97$_b$	0.278
C18:2n-6	7.75$_a$	0.201	11.26$_b$	0.316	33.07*$_a$	0.383	35.73*$_b$	0.430
C18:3n-3	0.44$_a$	0.017	0.67$_b$	0.020	0.50***$_a$	0.020	0.61***$_b$	0.025
C20:0	0.20$_a$	0.005	0.17$_b$	0.005	0.06	0.009	0.06	0.008
C18:3n-6	0.90$_a$	0.022	0.79$_b$	0.018	0.25	0.017	0.23**	0.010
C20:2n-6	0.37$_a$	0.016	0.47$_b$	0.021	0.47***	0.033	0.53***	0.039
C20:4n-6	0.18$_a$	0.009	0.27$_b$	0.012	10.34**$_a$	0.324	12.01**$_b$	0.416
C22:0	n.d.		n.d.		0.18	0.033	0.17	0.035
C20:3n-6	0.08$_a$	0.006	0.11*$_b$	0.011	0.67*	0.077	0.59***	0.060
C24:0	0.06*$_a$	0.004	0.08$_b$	0.004	0.67*	0.055	0.76	0.059
SFA	40.22$_a$	0.352	42.40$_b$	0.361	37.03	0.350	36.48	0.353
UFA	59.78$_a$	0.352	57.60$_b$	0.361	62.97*	0.350	63.52*	0.353
MUFA	50.06*$_a$	0.447	44.03**$_b$	0.465	17.66$_a$	0.364	13.83$_b$	0.300
PUFA	9.72$_a$	0.228	13.57$_b$	0.348	45.31*$_a$	0.530	49.69*$_b$	0.395

LD: *Longissimus dorsi*; *PMM*: *Psoas maior* and *minor.* SFA: sum of saturated fatty acids; UFA: sum of unsaturated fatty acids; PUFA: sum of polyunsaturated fatty acids; MUFA: sum of monounsaturated fatty acids.
[a,b]Means within the same row and lipid class not followed by the same number differ significantly (P<0.05). Significantly diferent values as influenced by diet *(P<0.05); **(P<0.01); ***(P<0.001).

The fattening diet clearly affects the fatty acid composition of NL and PL in both muscles and liver. (Table 3). The oleic acid (C18:1 n-9) content in NL and linoleic (C18:2 n-6) and linolenic (C18:3 n-3) acid contents in PL showed significant higher values in the LD and PMM muscles from pigs fed only with chestnuts. In these muscles, also the linolenic (C18:3 n-3) and arachidonic (C20:4 n-6) acids content in the PL was affected by diet. No significant differences were observed in SFA composition associated to the type of finishing diet.

Table 3. Fatty acid composition, % and standard error of the mean (SEM), of neutral and polar lipids in liver

	Neutral lipids (NL)		Polar lipids (PL)	
	Mean	**SEM**	**Mean**	**SEM**
C12:0	n.d.		n.d.	
C14:0	0.40	0.031	0.40	0.029
C14:1	0.03	0.006	0.09	0.006
C16:0	15.06	0.412	20.44	0.419
C16:1	0.95*	0.057	0.96	0.047
C17:0	0.45	0.031	0.60	0.045
C17:1	0.18	0.009	0.19	0.013
C18:0	18.55***	0.411	28.60***	0.803
C18:1n-9	25.89***	0.529	13.85***	0.652
C18:2n-6	17.99	0.332	17.62	0.363
C18:3n-3	0.80	0.036	0.57*	0.034
C20:0	0.04	0.003	0.04	0.010
C18:3n-6	0.33	0.014	0.18	0.022
C20:2n-6	0.42	0.021	0.58	0.033
C20:4n-6	16.59***	0.655	13.93	0.407
C20:3n-6	0.72	0.026	0.46	0.022
C24:0	1.58	0.106	1.41	0.117
SFA	36.09**	0.492	51.58***	0.693
UFA	63.91**	0.492	48.42***	0.693
MUFA	27.06***	0.569	15.10***	0.644
PUFA	36.85***	0.796	33.33	0.615

SFA: sum of saturated fatty acids; UFA: sum of unsaturated fatty acids; PUFA: sum of polyunsaturated fatty acids; MUFA: sum of monounsaturated fatty acids.
Significantly diferent values as influenced by diet *(P<0.05); **(P<0.01); ***(P<0.001).

It has been proposed that liver fatty acid composition may be a good indicator of feeding regime received by the pigs during the last fattening days (Ruiz *et al*., 1998). In this experiment was observed that inclusion of chestnuts in the diet significantly affected the palmitic (C16:0), stearic (C18:0) and oleic (C18:1 n-9) acid contents in both lipid classes in liver; the arachidonic (C20:4 n-6) and linolenic (C18:3 n-3) acid contents were also affected in NL and PL, respectively.

The factors associated with feeding that modify the composition in fatty acids of the tissues of the pig are: the amount of fat, proteins and carbohydrates present in the diet, the composition in fatty acids of the meal and the duration of the fattening period (Cava and Andrés, 2001).

Oleic acid (C18:1 n-9), is the most abundant fatty acid in NL of fat intramuscular and liver. The results show that the chestnuts inclusion increased the deposition of oleic acid (C18:1 n-9) in NL of both muscles and in NL and PL of liver. Doran *et al*. (2006) and Teye *et al*. (2006) showed that low protein diets increased the expression of stearoyl Co-A desaturase in *Longissimus dorsi* muscle and that there is a linear relationship between the expression of stearoyl Co-A desaturase and the amount of oleic acid (C18:1 n-9) in muscle. The protein content is lower in chestnuts than in the commercial feed compound, which would explain in part the high deposition of oleic acid (C18:1 n-9) in LN, fundamentally.

The linoleic (C18:2 n-6) and linolenic (C18:3 n-3) acids content of porcine fat is related to the linoleic (C18:2 n-6) and linolenic (C18:3 n-3) acids of the diet. The chestnuts have higher levels of linoleic (C18:2 n-6) and linolenic (C18:3 n-3) acids that commercial compound (Table 1). We found increased level of linoleic (C18:2 n-6) and linolenic (C18:3 n-3) acids in PL of LD and PMM muscles. Enser *et al*. (2000) showed that an increased dietary level of linoleic (C18:2 n-6) and linolenic (C18:3 n-3) acids results in an increased level of these fatty acids in both neutral and polar lipids of intramuscular fat. In other hand, the triglyceride content has a low fatty acid turnover rate and metabolic activity, average triglyceride life has been estimated to be over 180 days (Cunningham, 1968), a longer time than fattening phase of Celta pigs. Consequently, the concentration of the linoleic (C18:2 n-6) and linolenic (C18:3 n-3) acids does not accurately reflect the ingestion of chestnuts during the finish-fattening phase in NL.

The biosynthesis of arachidonic acid (C20:4 n-6) involves the desaturation and elongation of the dietary linoleic acid (C18:2 n-6) (Valette *et al*., 1991; Pérez-Palacios *et al*., 2009). The chestnuts inclusion in diet significantly affected the content of arachidonic acid (C20:4 n-6) on the PL of both muscles ($P<0.01$) and in the NL of liver ($P<0.001$).

The C20:2 n-6 and C20:3 n-6 acids were also affected on the PL of both muscles by the inclusion of chestnuts in the diet. These fatty acids derived from dietary linolenic acid (C18:3 n-3) and are deposited in muscle phospholipids but not in muscle neutral lipid (Enser *et al*., 2000), as shown in the present study.

IV – Conclusions

Metabolic type of muscle influences fatty acid composition of NL and PL in swine, those muscles with a predominantly oxidative metabolism showing higher amounts of PUFA in both lipid classes. On the other hand, we can observe a high response in fatty acid composition of in intramuscular and hepatic fat, as a result of chestnuts inclusion in the diet during the finish-fattening period.

Acknowledgments

This work was financially supported by the Spanish Ministry of Science and Innovation (Grant AGL2008-05274-C02-01/ALI). The authors also wish to thank to the Instituto Orensano de Desarrollo (INORDE) for its valuable collaboration in the rearing of the pigs.

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Natural antioxidant in pig feeding: Effects on meat and on salami quality

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Abstract. The purpose of the present work was to evaluate the influence of polyphenols supplementation of diet during the last period of fattening (30 days) on oxidative process and chemical composition of pork and dry sausage. Material and methods. At slaughtering 36 samples of *Biceps femoris* (BF) muscle from heavy pigs fed control diet or supplemented diet with a natural extract of Verbenaceae (Ajute^{TM}) were collected by each dietary treatment group (n=18). Salami Cremona (PGI) were produced by a local plant according to the Disciplinary of Production and 6 salami for each treatment were submitted to sensory evaluation. Results and conclusion. No differences were found for proximate composition neither in fresh meat nor in salami. The oxidative stability, assessed using thiobarbituric acid reactive substances (TBARS), is lower in fresh meat (P = 0.04) and in salami (P > 0.05) of treated group. Sensory evaluation of Cremona salami discriminated the muscle color (P = 0.04) and the sour taste descriptors (P = 0.02) that resulted more intense in the treated group. The use of natural extracts determined a longer shelf life of meat due to the lower oxidation and a greater colour stability of salami over three months of ripening.

Keywords. Natural antioxidant – Heavy pig – Meat quality – Salame.

Effet de l'ajout d'antioxydants végétaux dans l'alimentation des porcs sur la qualité des viandes et des saucissons

Résumé. *Deux groupes de 18 porcs (poids initial: 128 kg, poids final: 160 kg) ont été nourris avec un régime témoin ou supplémenté avec un antioxydant d'origine végétale (AOV). Des échantillons de viande (muscle* Biceps femoris *– BF) et de saucissons issus de ces porcs ont été analysés: composition chimique, mesure des potentiels de lipopéroxydation (TBARS) et analyse sensorielle pour les saucissons. Les résultats montrent que l'ajout d'AOV dans les rations permet de diminuer les teneurs en TBARS dans le muscle BF. L'analyse sensorielle des saucissons secs montre que l'ajout d'AOV n'a pas d'effets majeurs sur les qualités organoleptiques (effet significatif limité sur la couleur et l'acidité).*

Mots-clés. *Antioxydants végétaux – Porc lourd – Qualité de la viande – Salami.*

I – Introduction

Lipid oxidation is a major cause of chemical spoilage in food systems. To avoid or delay this autoxidation process, antioxidants such as Vitamin E in animal feeding have been utilised with the practice successfully in order to improve meat quality (Corino *et al*., 1999 a,b) and processed pork products (Zanardi *et al*.,1999). Natural antioxidant have been widely reported potent antioxidant effects (Nakatani 2000). The antioxidant properties are related especially to their phenolic contents. Among the polyphenols, verbascoside, constituent of many plants for food, flavoring and pharmaceuticals shows the highest scavenger activity inside the PPG tested (Wang *et al*., 1996). Recently Rossi *et al*., (2009) found that verbascoside has greater antioxidant power compared to other phenolic compounds and Trolox. The effects of phytochemical antioxidants on lipid oxidation in meat and meat products is of major concern. Considering the antioxidant effect of verbascoside tested, we likely hypothesized on the basis of a our preliminary study (Corino *et al*., 2007) that it could influence meat and processed meat quality. The aim of the present work was to evaluate the influence of polyphenols supplementation of diet in pigs during the last period of fattening (45 days) on qualitative characteristics of pork and dry sausage.

II – Materials and methods

Hybrid Dalland pigs (36 female) of average weight 128 kg ± 13.2 were divided into two groups of 18 animals. The groups were randomly assigned to a growing-finishing diet supplemented with a natural extract of Verbenacee, titered in polyphenols, expressed as verbascoside, as to apport 6 mg/kg of feed (TR) or not (CON). Daily feed intake was restricted to 9% of metabolic live with water available for *ad libitum* consumption. After 45 d of feeding period, the animals were slaughtered at a commercial slaughter house (average weight of 160 kg). *Biceps femoris* (BF) muscle was sampled in order to analyze chemical composition (AOAC, 2000), drip loss (Rasmussen *et al*., 1996), cooking loss (Honikel 1998) and lipid oxidation measured as TBARS (Monin *et al*., 2003). Moreover two batches (TR and CON) were produced and sent to a processing factory in order to produce Salame Cremona (PGI) according to the Disciplinary of Production. Salame Cremona is an uncooked cured and matured sausage product Commission Regulation (EC) No 1362/2007. Six salami for each treatment were submitted to stability oxidation, chemical composition, color coordinates evaluation (L*, a*, b*) recorded using a Chroma Meter CR-300 (Minolta Cameras, Osaka, Japan) and sensory evaluation. Determinations were made in duplicate. Sensory analysis was performed using sensory profile method (ISO 13299, 2003). Descriptors selected were: muscle and fat colour, cohesion thin/fat, homogeneity of colour, consistency, oily, easily to peel, rancid, acid, salt, sweet and aged flavour. Samples were rated on a linear scale ranged from 1 (=minimum intensity) to 9 (=maximum intensity). All data were analysed by "Esplora" procedure in order to find outliers values, tested to the normality distribution and analyzed by T Student test with treatment as fixed effect (SPSS Inc., Chicago, IL).

III – Results and discussion

Results on chemical composition of fresh muscle and salame Cremona are presented in Table 1. No significant difference were found in any parameters analyzed. Results on chemical composition of muscle are in agreement with data reported in heavy pigs (Corino *et al*., 2002; Ventanas *et al*., 2006) and in BF muscle (Franci *et al*., 1995). The values found in Salame Cremona are in agreement with the Disciplinary of Production that establish a value of crude protein higher than 20% and water protein ratio maximum 2. Dietary treatment did not modify drip loss, according with Correa *et al*. (2006) on growing pigs.

Table 1. Chemical composition and drip loss of *Biceps femoris* muscle and Salame Cremona†

Item	BF muscle		Effect	Salame Cremona		Effect
	CON	TR	P	CON	TR	P
Moisture	77.73	77.56	NS	31.88	31.87	NS
Crude protein	22.26	22.43	NS	23.81	24.01	NS
Ether extract	3.82	3.30	NS	30.54	30.07	NS
Ash	1.12	1.24	NS	5.60	5.99	NS
Drip loss	4.14	4.34	NS	-	-	NS
Cooking loss	14.95	14.50	0.12	-	.	-

†Data expressed as percentage of wet weight.

The National Pork Board suggest a drip loss values equal to 2.5% at 24 h as indicator of meat quality; anyway the muscle analysed in the present work is different from that one individuated from the Commission. Cooking loss is a combination of liquid and soluble matters lost from the meat during cooking. The water is probably lost due to heat induced protein denaturation during cooking of the meat, which causes less water to be entrapped within the protein structures held by capillary forces. In this study even if no significant effect was deemed (P = 0.125), the treated group showed a numerically higher (+ 3%) retention value, important trait for the quality of pork meat since it is consumed especially cooked and therefore subjected to a protein denaturation. Verbascoside supplementation positively influenced the oxidative stability of fresh muscle (P<0.05) and salame Cremona (P>0.05) (Fig. 1). The effect is likely due to the antioxidant properties of the natural substances used and/or to a "saving" action on the Vitamin E as demonstrated by Casamassima *et al*., (2009). Studies on natural substances and their extracts have sometimes inconsistent results. Janz *et al*., (2007) did not find any effect on oxidative stability of pork meat from pigs fed with 0.05 % of essential oil, garlic, origanum and ginger. On the contrary Mason *et al*., (2005) reported that a supplementation with green tea, cathechin (200 mg/kg) in pigs from weaning to slaughtering improved significantly the oxidative stability of the LD muscle. The TBARS values of BF muscle are lower than those reported in literature; the reason could be dependent on the type of muscle used. In fact Novelli *et al*., (1998) analysed different muscles and found a value equal to 0.27 mg/kg MDA in BF muscle.

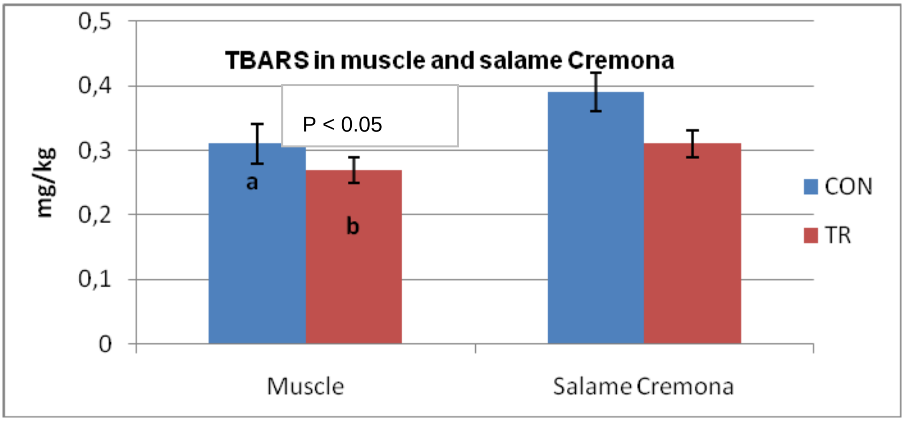

Fig 1. TBAR on BF muscle and on Salame *Cremona.*

In the present work the treated group showed values similar to those found by McCarthy *et al*., (2001) who reported 0.25 mg/kg of TBARS in BF samples muscle from pigs fed diets supplemented with different natural substances such as rosemary, aloe, ginseng, tea. The percentage reduction found herein is similar to that found by Corino *et al*. (1999), in LD muscle from pigs fed with a high Vitamin E dose (300 mg/kg). Concerning the transformed product, Martinez *et al*., (2006) found lower (P<0.05) values of TBARS in sausage treated with increasing values of green tea and borage. Results on Salame Cremona are in agreement with those found by Novelli *et al*., (1998) and Zanardi *et al*., (1998) in salame Milano in which values range from 0.22 to 0.30 mg malonaldehyde/kg sample. No significant difference was found for any descriptor excepting for muscle color and acid more intense in the treated group than CON group; this result is consistent with strumental data analysis (data not shown). Anyway sensorial analyses, for acid descriptor, releaved a values equal to one, far away to the maximum intensity fixed to nine. The result on colour agrees well with Ventanas *et al*., (2007) confirming the stability of the color during the seasoning process.

IV – Conclusions

In the present work a positive effect of dietary natural antioxidant has been noted, showing an improved oxidative stability equal to +15% in raw meat. Moreover the unchanged chemical characteristics of meat and salame Cremona and the surface color of salame stabilized also after a ripening period indicate that polyphenols supplementation in pig feeding is an effective way to improve color and lipid stability of muscle and transformed product.

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Ultrasonographic *in vivo* estimation of back fat depth and *Longissimus dorsi* area in Iberian pigs

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Abstract. Real-time ultrasound is a useful technology for measuring carcass composition in live animals and to select future seedstock for carcass merit. Therefore, the objective of this study was to evaluate the use of ultrasound to predict the amount of fat and muscle in Iberian pigs. A total of 163 Iberian pigs were ultrasonically scanned one day before slaughter, and carcass composition was also determined in the slaughterhouse. Ultrasound and carcass traits included the three lays of back fat and *Longissimus dorsi* (LD) area, at the 10^{th} and 14^{th} rib levels. Data were analyzed by regression and correlation procedures using SAS. Results indicated a positive correlation between ultrasound and carcass traits at both rib levels. Regression results indicated that ultrasound measurements had smaller values than carcass measurements. Correlations between ultrasound back fat measurements and carcass, ham, and foreleg weights were positive, but they were negative with respect to LD weight. The highest correlation with LD weight was LD area at the 14^{th} rib level.

Keywords. Ultrasound – Iberian pigs – Back fat – *Longissimus dorsi.*

Estimation échographique* in vivo *de l'épaisseur de la graisse dorsale et de la surface du* Longissimus dorsi *chez les porcs Ibériques

Résumé. *Les ultrasons sont un outil valide et économique pour mesurer la composition de la carcasse des animaux vivants et sélectionner les futures reproductrices. Ce travail évalue l'utilisation des ultrasons pour prédire la quantité de tissu gras et musculaire chez le porc Ibérique. 163 porcs Ibériques ont été échographiés un jour avant l'abattage, et ultérieurement on a étudié leurs carcasses à l'abattoir. Par ultrasonographie, et ultérieurement sur la carcasse, on a mesuré l'épaisseur des trois couches de graisse dorsale et la surface du* Longissimus dorsi *(LD), au niveau du dixième et du quatorzième espace intercostal. Les données ont été analysées par des procédures de régression et corrélation de SAS. Les résultats obtenus montrent une corrélation positive entre les mesures par ultrasons et celles de la carcasse au niveau de la dixième et la quatorzième côte. Les résultats de la régression montrent que les mesures ultrasoniques sont sous-estimées par rapport à celles de la carcasse. Les corrélations entre les mesures ultrasoniques de la graisse dorsale par rapport au poids de la carcasse, au jambon, et à l'épaule étaient positives; mais elles étaient négatives par rapport au poids du filet. La mesure la plus corrélée au poids du filet a été la surface du filet dans la quatorzième côte.*

Mots-clés. *Ultrason – Porc Ibérique – Graisse dorsale – Longissimus dorsi.*

I – Introduction

The Iberian pig breed has a traditional production system linked to natural resources, since pigs are finished grazing on pastures and acorns. This system produces meat with a high percentage of intramuscular fat and a high proportion of monounsaturated fatty acids (Ventanas *et al*., 2006), thus providing excellent sensorial and healthy properties to the cured products, which are the source of a thriving meat industry and a renowned quality certification. However, to reach this high percentage of intramuscular fat, animals also deposit a large amount of fat (around 90 mm thick). There is an important genetic variation in swine for fat deposition traits, thus enabling the possibility of selecting superior individuals for intramuscular fat with lower levels of subcutaneous fat (Newcom *et al*., 2005). However, measuring carcass traits in live animals for selection purposes is not an easy task. Ultrasonography has been used to develop predictive equations for estimating intramuscular fat in live cattle (Wilson *et al*., 2001), and in pigs (Newcom *et al*., 2002). *In vivo* ultrasound estimates for loin area and fat thickness in Duroc pigs were very reliable (Newcom *et al*., 2002, 2005). The objective of this study was to assess the capabilities of ultrasound for measuring back fat thickness and loin area in the Iberian pig in order to develop a predictive method for meat yield, thus relating variables measured with ultrasound in the live animal with those taken in the carcass after slaughter.

II – Materials and methods

Animals: For this study, 163 Iberian pigs of the red *("Retinto"*) strain were used. Animals were raised indoors in standard commercial conditions up to 25 Kg of body weight (BW). After this period, animals were raised outdoors in extensive conditions and fed a concentrate in a restricted diet (as usual in this traditional system) up to 12 months of age. At this age, animals were divided in five groups and started an "ad libitum" feeding period with different types of food: One group was fed only pasture and acorn; the second was fed pasture and acorn up to 135 kg of BW and then finished with a standard, commercial concentrate; the third group was fed pastures, acorn and standard concentrate at the same time; the forth group was fed standard concentrate; and the fifth group was fed a commercial concentrate having a high level of oleic acid to imitate acorn composition. Animals were fed for a period of 2.5 to 3 months and were slaughtered after reaching 135 kg of BW (135 to 175 kg range). To allow for a thorough study of carcasses, no more than 20 animals at a time were slaughtered.

Ultrasound image collection: One day before slaughter, pigs were ultrasonically scanned to measure body composition, using an Aloka 500 apparatus (Aloka Holding – Europe, Switzerland) and a 3,5 MHz, 12 cm long probe. Ultrasound images were collected by placing the probe perpendicular to the loin a two different rib levels: one image was taken between the 10th and 11th ribs (10th intercostal space), and the other was taken just behind the last (14th) rib. A soft, rubbery adaptor made of Superflap® was used between the animal and the probe to allow for an adequate contact despite of the curved back surface. Images were digitalized and stored in a computer. Image measurements were done afterwards by using the Biosoft® software (Biotronics inc., Ames, IA, USA). For each image, loin area and thickness of the 3 backfat (BF) layers were measured. The ultrasound ("u") measurements, taken at the 10th intercostal space and behind the last rib, respectively ("10", "14"), were the following: external ("E") back fat layer (EBFu10 and EBFu14); middle ("M") layer (MBFu10 and MBFu14); internal ("I") layer (IBFu10 and IBFu14); external + middle ("EM") layers (EMBFu10 and EMBFu14); and total ("T") backfat thickness (BFu10 and BFu14).

Carcass data: After slaughter, carcass weight (CW) and weight and yields of commercial cuts (ham, foreleg and loin) were measured. In addition, a portion of loin containing 4 chops (spanning from the 11^{th} to the14^{th} ribs) was extracted from the carcass and used to measure back fat layers thickness and loin area at the two already mentioned rib levels.

Variables were analyzed by using correlation and regression procedures in SAS.

III – Results and discussion

The correlation coefficients between the ultrasound variables and the corresponding carcass variables at the two rib levels are presented in Table 1. The variables with a higher correlation were middle BF layer (r = 0.72 and 0,61) and total BF (r = 0.67 and 0,66) for the 10th intercostal space ("10") and last space ("14"), respectively. Prior estimates of BF10 gave correlations of 0.55 (McLaren *et al*., 1989) and 0.93 (Turlington, 1992). The correlation coefficient between ultrasound and carcass loin area was 0.36 and 0.41 for the 10-11 intercostal space and last rib space, respectively. This correlations are similar to those reported by Daza *et al*. (2006) in Iberian pigs. Correlations reported by Moeller (1998) and Lopes *et al*.

(1987) range between 0.27 and 0.73. In general, the correlations from our study are slightly lower than those from the literature, maybe because larger slaughter weights result in these correlation being lower (Daza *et al*., 2006). In addition, we found in our study that Iberian pigs show a wide variation in loin section shape, so that the internal fat layer is sometimes hard to measure as accurately as expected *in vivo* and in the carcass. Nevertheless, considering that carcass measurements also have an associated error, the magnitude of the correlations indicate that ultrasonography can be a good alternative to measure live animals for selection purposes, as indicated by Moeller *et al*. (1998).

Table 1. Correlations between ultrasound and carcass measurements in the two anatomical locations

Rib level	EBFuc	MBFuc	IBFuc	EMBFuc	Bfuc	Aluc
10th	0.56	0.72	0.39	0.72	0.67	0.36
14th	0.27	0.61	0.33	0.60	0.66	0.41

Backfat layers: EBFuc=external; MBFuc=middle; IMBFuc=internal; EMBFuc=external+medial; BFuc=total backfat. ALuc=*Longissimus dorsi* area.

Tables 2 and 3 depict, for the two rib levels, the correlations between ultrasound ("u") or carcass ("c") measurements and the weights and yields of meat cuts, namely Foreleg Weight (FW), Ham Weight (HW), Loin Weight (LW), Foreleg Yield (FY), Ham Yield (HY) and loin yield (LY).

Table 2. Correlations between ultrasound measurements (u) and the carcass, foreleg, ham and loin weights and yields

	EBFu10	MBFu10	IBFu10	BFu10	Alu10	EBFu14	MBFu14	IBFu14	BFu14	Alu14
CW	0.25	0.38	0.29	0.43	0.15	0.29	0.27	0.22	0.33	0.21
FW	-0.03	0.03	0.09	0.04	0.23	0.13	-0.08	-0.01	-0.03	0.18
HW	0.24	0.09	0.03	0.13	0.24	0.21	0.10	-0.07	0.07	0.23
LW	-0.15	-0.44	-0.34	-0.47	0.36	-0.09	-0.36	-0.26	-0.37	0.43
FY	0.05	-0.33	-0.30	-0.33	0.15	-0.05	-0.19	-0.37	-0.30	0.08
HY	-0.30	-0.38	-0.21	-0.42	0.11	-0.15	-0.40	-0.26	-0.41	0.00
LY	-0.23	-0.53	-0.42	-0.58	0.20	-0.20	-0.41	-0.31	-0.45	0.25

Backfat layers: EBFu=external; MBFu=middle; IMBFu=internal; EMBFu=external+medial; BFu=total backfat. ALu=*Longissimus dorsi* area. CW=carcass weight; FW= foreleg weight; HW= ham weight; LW= loin weight; FY= foreleg yield; HY= ham yield; LY= loin yield.

Table 3. Correlations between carcass measurements (c) and the carcass, foreleg, ham and loin weights and yields

	EBFc10	MBFc10	IBFc10	BFc10	Alc10	EBFc14	MBFc14	IBFc14	BFc14	Alc14
CW	0.40	0.44	0.40	0.59	-0.10	0.15	0.49	0.06	0.46	-0.04
FW	0.05	0.14	0.22	0.21	0.05	0.10	0.11	-0.08	0.06	0.11
HW	0.39	0.18	0.19	0.31	0.03	0.20	0.26	-0.17	0.13	0.02
LW	-0.20	-0.46	-0.35	-0.53	0.64	-0.03	-0.39	-0.022	-0.46	0.58
FY	0.08	-0.26	-0.19	-0.24	0.16	0.11	-0.21	-0.32	-0.35	0.06
HY	-0.034	-0.29	-0.16	-0.36	0.17	-0.03	-0.36	-0.16	-0.40	0.18
LY	-0.32	-0.58	-0.47	-0.69	0.58	-0.08	-0.53	-0.21	-0.58	0.49

Backfat layers: EBFuc=external; MBFuc=middle; IMBFuc=internal; EMBFuc=external+medial; BFuc=total backfat. ALuc=Longissimus dorsi area. CW=carcass weight; FW= foreleg weight; HW= ham weight; LW= loin weight; FY= foreleg yield; HY= ham yield; LY= loin yield.

Any of the measurements have a correlation above 0.4 for FW, HW, and FY. In particular, LW had correlations above 0.40 with MBFu10, BFu10, and ultrasound loin area-14. In addition, HY has correlations above 0.4 MBFu14, BFu10 and BFU14 and LY with MBFu10, MBFu14, BFu10, BFu14 and IBFu10. Carcass measurements had larger correlations than ultrasound measurements, specially for loin area, contrarily to results reported by Daza *et al*. (2006). It can be concluded that ultrasound backfat measurements have similar prediction abilities than carcass backfat, but this was not the case for ultrasound loin areas.

Table 4 describes the statistics of individual regressions between carcass and ultrasound variables, which serve to evaluate the prediction ability and the possible bias of each ultrasound variable. Together with high values of R2, the closest the intercept to zero and the slope to 1, the better the prediction ability. Regression resulted as expected, thus corroborating the correlation results. In addition, these results indicate that ultrasound loin areas and total backfat underestimated carcass loin areas and total backfat. However, it is important to consider that carcass measurements also carried measurement errors.

Table 4. Regression of carcass to ultrasonic measurements for backfat layer variables and loin area at two anatomical locations (10th and 14th rib levels).

VARIABLES	N	R^2	Intercept	Slope
EBFu10	156	0.31	0.84	0.72
EMBFu10	139	0.63	1.97	0.82
BFu10	156	0.51	2.32	0.77
ALu10	118	0.35	4.22	0.82
EBFu14	153	0.07	0.96	0.43
EMBFu14	152	0.35	2.09	0.69
BFu14	141	0.56	2.92	0.72
ALu14	119	0.22	9.7	0.91

Backfat layers: EBFu=external; MBFu=middle; IMBFu=internal; EMBFu=external+medial; BFu=total backfat. ALu=*Longissimus dorsi* area.

In conclusion, the results of this study are in agreement to results reported by other authors working with Iberian pigs. Ultrasound technology seems a good tool to measure live Iberian pigs. However, considering the heavy slaughter weights, the large backfat thickness and the loin shape variability shown by Iberian pigs, this technology needs to be readjusted to increase its accuracy for this breed.

Acknowledgements

This study was financed by FEDER (PDT09B039), INIA (RTA2007-0093-00-00) and Junta de Extremadura.

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Acidic profile in two different muscles of Nero Siciliano pigs as affected by different finishing diets

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Abstract. Fat and fatty acids contribute significantly and in various ways to meat quality and are central to the nutritional value of meat. Meat quality is also influenced by intrinsic factors such as muscle type. Therefore, the study examined the fatty acid composition of the white (*Longissimus dorsi* - LD) and the red (*Psoas major* - PM) muscle from 24 Nero Siciliano pigs fed with two different diets. The trial was carried out in the Nebrodi mountain area of Sicily (Italy). During the fattening period, the animals were divided into two groups, kept in two distinct wooded areas of 6 hectares each, and fed exclusively on acorn (A) and germinated barley (B). Results showed a strong interaction between diet and muscle type. The PUFA to SFA ratio was higher in PM muscle for both diets with A 0.16 LD *vs* 0.25 PM ($P \leq 0.0001$) and B 0.16 LD *vs* 0.21 PM ($P \leq 0.001$). The C18:2 n-6 amount, entirely derived from the diet, showed high statistical significance both as dietary effect and as intrinsic (muscle-type related) factor. In order to further improve strategies for pork production, more information on the effects of inherent factors (e.g. muscle type) on meat traits is needed.

Keywords. Nero Siciliano pig – Meat quality – Fatty acids.

Évaluation de l'effet de deux diètes sur le profil en acides gras de deux muscles différents chez le porc Nero Siciliano

Résumé. *Les composantes lipidique et acide jouent un rôle central dans l'évaluation nutritionnelle de la viande porcine, en contribuant à la détermination de la qualité, qui est influencée aussi par des facteurs intrinsèques à l'animal, dont le type de muscle. Pour cette raison, cette étude a évalué le profil en acides gras du muscle blanc (*Longissimus dorsi *- LD) et rouge (*Psoas maior *- PM) de 24 porcs "Nero Siciliano" nourris avec deux régimes alimentaires différents au cours de la période de finition. L'essai a été effectué en Sicile (Italie), dans le parc des Nebrodi; les animaux ont été divisés en deux groupes (A et B) homogènes et gardés dans deux parcelles boisées différentes de 6 ha chacune. Le groupe A a été nourri exclusivement avec des glands et le groupe B avec de l'orge germée. Les résultats ont mis en évidence une forte interaction entre le régime alimentaire et les types de muscle examiné; le ratio PUFA/SFA a été plus élevé dans le muscle PM dans les 2 groupes: A 0,16 LD* vs *0,25 PM (P≤0,0001) et B 0,16 LD* vs *0,21 PM (P≤0,001). La quantité de C18:2 n-6, dérivée de la ration, a été significativement corrélée à la ration et au type musculaire. Il est nécessaire d'approfondir pour définir de nouvelles stratégies de production.*

Mots-clés. *Porc Nero Siciliano – Qualité de la viande – Acides gras.*

I – Introduction

The susceptibility of pig muscles to lipid oxidation and the acidic profile in intramuscular fat depends on various factors, e.g. fatty acid composition of cell membranes, balance between antioxidants and prooxidants, amount and composition of lipids, and the activity of certain enzymes (Lauridsen *et al.,* 1999; Andrés *et al*., 2001). Numerous studies have demonstrated the correlation between diet, muscle type and acidic profile of meat. (Wood *et al*., 2004; Chang *et al*., 2003). It is also known that the fatty acid composition and the concentration of fatty acids can influence the types and the concentration of volatiles produced in heated fat and dry-cured products (Andrés *et al.*, 2001). The aim of this study was to examine the effect of the diet and the influence of two muscles with different (oxidative or glycolytic) metabolism on the fatty acid composition of intramuscular fat of Nero Siciliano pigs, an autochthonous Italian breed reared in a traditional, free-range production system and fed on acorn and grass.

II – Materials and methods

Twenty-four "Nero Siciliano" pigs were in outdoors reared in the Nebrodi mountain region of Sicily. Animals were assigned to two groups called Acorn (A) and Barley (B), consisting of 12 animals each, homogenous for sex (castrated males) and body weight (BW, 79.48 ± 0.15 kg). Animals of group A were kept in a wooded area of 6 hectares, appropriately enclosed, and fed with acorn during the fattening period (90 days). Animals of group B were reared within an open-air system in the same rural region and fed with germinated barley on a basis of 2.5 kg/pig/d during the fattening period. After 90 days and a fasting period of 18 hours (ASPA, 1991), animals were slaughtered. A sample of *Psoas major* (PM) and *Longissimus dorsi* (LD) muscle tissue was taken from each of the 24 carcasses. Each sample was examined for its crude fat (AOAC, 2005), followed by an analysis of its acidic composition. The fatty acid composition was determined on lipids extracted by an automatic extractor Foss Model Soxtec. Fatty acids methyl esters of the intramuscular fat were prepared by direct transesterification with sulphuric acid : methanol 1:2 (Christie, 1993) and analysed using an Agilent Technologies 6890N (U.S.A) gascromatograph operated with a fused silica capillary column OMEGAWAX 250 (Supelco, U.S.A.), 30m x 0.25mm I.D., 0.25 μm film thickness. Column temperature was programmed: initial isotherm of 160°C (6 min.), increment of 3°C/min and final isotherm of 250°C (30 min.). Carrier gas: helium (1 m L/min). Peak areas were expressed in percentage of the total fatty acid identified. On the basis of the fatty acid identified, the quality indices were calculated using the equations proposed by Ulbricht and Southgate (1991). Diets (acorn or germinated-barley), muscles (LD or PM) and their interaction were compared by a two-way ANOVA, using the GLM procedure of SAS (2001).

III – Results and conclusions

The FA composition of *Psoas major* (PM) and *Longissimus dorsi* (LD) muscle is shown in Table 1. The two experimental groups showed statistical differences for most of the FA detected. However, the diet did not produce a consistent effect on the proportions of some FA in the tissues examined. In the Acorn group (A) total saturated FA (SFA) was found to be higher in the LD muscle, whereas in the Barley group (B) SFA was higher in the PM muscle. The levels of stearic acid (C18:0) showed high statistical differences for the LD muscle ($P \leq 0.0001$) (12.75% *vs* 8.40% in A and B, respectively) and no statistical differences for the PM muscle (13.23% *vs* 13.66% in A and B, respectively). These different results could be explained by the fact that a high proportion of SFA comes from de novo synthesis, and only a small proportion of total SFA is directly accumulated from dietary fatty acids (Monahan, *et al.*, 1992). In the B, total monounsaturated FA (MUFA) was higher in both muscles. In the A, the level of oleic acid (C18:1n-9) showed a high statistical difference for the LD muscle ($P \leq 0.0001$) (48.23% *vs* 37.14% in A and B, respectively), whereas in the B a statistical difference was found for the PM muscle ($P \leq 0.01$) (39.57% *vs* 43.06% in A and B, respectively). The apparently different influence of the diet on the oleic acid (C18:1 n-9) content in the muscles examined in this work could be explained by the reported effects of muscle fibre types (Andrés *et al.*, 1999; Muriel *et al.,* 2002). In the A, the level of PUFA was higher in both muscles. In detail, the content of PUFA in the LD muscle was ($P = 0.03$) 6.09% *vs* 5.24% in A and B and the content of PUFA in the PM muscle resulted ($P= 0.001$) 9,49% *vs* 8.16% in A and B respectively. These differences might be cleared by the high content of poliphenols in the acorn, reducing the oxidation of lipids. However, neutral lipids from the oxidative muscle PM contained higher percentages of linoleic acid (C18:2 n-6), (7.11 in PM *vs* 4.77 in LD for the A and 6.19 in PM *vs* 4.01 in LD for the B respectively) and arachidonic acid (C20:4 n-6), (1.30 in PM *vs* 0.56 in LD for the A and 0.98 in PM *vs* 0.49 in LD for the B respectively).

Table 1. Principal fatty acid profiles (% methyl esters listed) of intramuscular fat of *Psoas major* (PM) and *Longissimus dorsi* (LD) muscles from Nero Siciliano pigs fed with different fattening diets

Fatty Acids	Muscle	Diet		P-value
		Acorn	Barley	
C14	LD	1.19	1.24	ns
	PM	1.23	1.17	ns
C16	LD	21.49	22.18	ns
	PM	22.87	23.74	***
C16:1	LD	3.93	4.61	***
	PM	3.44	3.50	ns
C18	LD	12.75	8.41	***
	PM	13.23	13.66	ns
C18:1 n-9	LD	48.23	37.14	***
	PM	39.58	43.06	**
C18:1 n-7	LD	4.31	19.05	***
	PM	7.82	4.61	*
C18:2 n-6	LD	4.77	4.01	**
	PM	7.11	6.19	**
C18:3 n-3	LD	0.24	0.26	ns
	PM	0.36	0.26	***
C20	LD	0.22	0.30	***
	PM	0.26	0.27	ns
C20:4 n-6	LD	0.56	0.49	ns
	PM	1.30	0.98	**
∑SFA	LD	36.22	32.47	***
	PM	38.42	39.51	ns
∑MUFA	LD	57.73	62.23	***
	PM	52.21	52.39	ns
∑PUFA	LD	6.09	5.25	*
	PM	9.50	8.16	**
∑PUFA n-3	LD	0.44	0.39	ns
	PM	0.61	0.53	*
∑PUFA n-6	LD	5.66	4.85	*
	PM	8.89	7.63	**
PUFA/SFA	LD	0.16	0.16	ns
	PM	0.25	0.21	**
AI	LD	0.41	0.40	ns
	PM	0.45	0.47	ns
TI	LD	1.07	0.92	***
	PM	1.16	1.22	ns

Results are expressed as percentage of total fatty acid methyl esters identified. AI, atherogenic index; TI, thrombogenic index; SFA, saturated fatty acids; MUFA, monounsaturated fatty acids; PUFA, polyunsaturated fatty acids); * $P \leq 0.05$; ** $P \leq 0.01$; *** $P \leq 0.0005$.

So, contrarily to Leseigneur and Gandemer (1991), the results of this study indicate a clear effect of the muscle type on the fatty acid profiles of neutral lipid fractions. In conclusion, muscle quality characteristics differed significantly according to their respective metabolic patterns. These variations are of interest because they might produce a different behaviour of the muscles during refrigeration display, freezing or culinary practices on the oxidative and lipolytic changes and their shelf-lives; therefore, these variations should not be ignored when studying the effects of feeding strategies on meat quality.

Acknowledgements

The authors wish to acknowledge the financial support received by MIUR- PRIN 2005 and by PRA 2005 (Prof. A. Zumbo).

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Influence of finishing diet on fatty acid profile in *Psoas major* muscle of Nero Siciliano pig

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Abstract. Skeletal muscle is a heterogeneous tissue composed of different fibre types, and differences in meat quality traits between "white" and "red" muscles and between certain breeds are well established. Therefore, the study examined the fatty acid composition of the red (*Psoas major* - PM) muscle in twenty-four Nero Siciliano pigs fed with two different diets. The trial was carried out in the Nebrodi mountain area of Sicily (Italy). During the fattening period, the animals were divided into two groups, kept in two distinct wooded areas of 6 hectares each, and fed exclusively on acorn (A) and germinated barley (B). Results showed that the concentrations of the long chain n-6 and n-3 fatty acids (which were recently found to play an important role in human nutrition) differed significantly between the two groups (P=0.015 for long chain n-6 and P=0.048 for long chain n-3). The linoleic acid (A 5.96 *vs* B 5.10; P=0.006) and α-linolenic acid (A 0.30 *vs* B 0.25; P=0.028) were higher in group A. Both group had high concentrations of C18:1 n–9, but in group A (43.27) concentrations were significantly (P=0.006) higher than in group B (40.10).

Keywords. Nero Siciliano pig – Acorn – Fatty acids – *Psoas*.

Influence de l'alimentation de finition sur le profil en acides gras du muscle* Psoas major *du porc Nero Siciliano

Résumé. *Le muscle squelettique est un tissu hétérogène comprenant divers types de fibres. Les différences existantes entre les muscles « blancs » et « rouges » et entre les races qui influencent les caractéristiques qualitatives de la viande sont bien connues. Le profil en acides gras du muscle rouge (*Psoas major *- PM) de 24 porcs de race Nero Siciliano alimentés avec deux rations différentes de finition a été examiné. L'épreuve a été réalisée en Sicile (Italie), dans le parc des «Nebrodi» ; les animaux ont été subdivisés en deux groupes homogènes et tenus dans deux parcelles boisées différentes de 6 ha chacune, dans lesquelles ils recevaient respectivement glands (A) et orge germée (B). Les résultats obtenus ont mis en évidence que les concentrations en acides gras à chaîne longue n-6 et n-3 (dont le rôle important en nutrition humaine a été vérifié récemment) sont significativement différentes entre les deux groupes (P=0,015 pour le n-6 et P=0,048 pour le n-3). Les concentrations en acide linolénique (A 5,96 vs B 5,10 ; P=0,006) et en acide α-linolénique (A 0,30 vs B 0,25 ; P=0,028) ont été plus élevées dans le groupe A. Les deux groupes (glands et orge germée) ont montré des teneurs élevées en C18:1n-9, mais pour A (43,27) elle a été significativement plus élevée (P=0,006) que pour B (40,10).*

Mots-clés. *Porc Nero Siciliano – Glands – Acides gras –* Psoas.

I – Introduction

The Nero Siciliano pig is a rustic pig breed reared in Sicily (Southern Italy). Compared with industrial pig production systems, the Nero Siciliano pig production process lasts longer. As to the diet, the final months before slaughter represent the crucial phase of the production process, because the feed characteristics (e.g. of acorn, grass or local concentrate feeds) during the critical finishing period significantly affect meat quality. In recent years, the consumers' interest in the so-called 'natural', 'bio' or 'organic' meats has been increasing. Therefore, meat from pig production systems in which pigs are free-range reared and fed on natural feeds with no growth promoters and antibiotics, has become an important field of interest. But the high quality of meat and meat products is the result of varoius factors, including genetics, rearing system and muscle type. Muscles are composed of different type of fibres (aW, aR and bR) with different contractile and metabolic properties (Cava *et al.*, 2003). Depending on the types of fibre that constitute a muscle, it differ in fat deposition, heme pigment concentration and phospholipid and fatty acid composition. Meats are more tasty and juicy, and total heme pigments and lipids oxidise faster in oxidative muscles than in glycolytic ones; this contributes to different textural properties (Wood, Wiseman and Cole,1994). The aim of this study was to examine the characteristics and the acidic profile of *Psoas major* muscle, a fast oxidative-glycolytic α-red muscle, in pigs fed with two different fattening diets.

II – Materials and methods

Twenty-four “Nero Siciliano” pigs were reared in outdoors in the Nebrodi mountain region of Sicily. Animals were assigned to two groups called Acorn (A) and Barley (B), consisting of 12 animals each, homogenous for sex (castrated males) and body weight (BW, 79.48 ± 0.15 kg). Animals of group A were kept in a wooded area of 6 hectares, appropriately enclosed, and fed with acorn during the fattening period (90 days). Animals of group B were reared within an open-air system in the same rural region and fed with germinated barley on a basis of 2.5 kg/pig/d during the fattening period. After 90 days and a fasting period of 18 hours (ASPA, 1991), animals were slaughtered. A sample of *Psoas major* (PM) muscle tissue was taken from each of the 24 carcasses. Each sample was examined for its crude fat (AOAC, 2005), followed by an analysis of its acidic composition. The fatty acid compositions were determined on lipids extracted by an automatic extractor Foss Model Soxtec. Fatty acids methyl esters of the intramuscular fat were prepared by direct transesterification with sulphuric acid : methanol 1:2 (Christie, 1993) and analysed using an Agilent Technologies 6890N (U.S.A) gaschromatograph operated with a split/splitless injector, a Gerstel autosampler MPS2 (Germany), a flame ionization detector and fused silica capillary column OMEGAWAX 250 (Supelco, U.S.A.), 30 m x 0.25 mm I.D., 0.25 μm film thickness. Column temperature was programmed: initial isotherm of 160 °C (6 min.), increment of 3°C/min and final isotherm of 250°C (30 min.). Temperature of the injector and detector: 250°C. Injection volume: 1.0 μ L. Carrier gas: helium (1 m L/min). Split ratio: 1:50. Identification of fatty acids was made by comparing the relative retention times of FAME peaks from samples with standards from Supelco (U.S.A.). Peak areas were acquired and calculated by Chemstation software (Agilent, U.S.A.) and expressed in percentage of the total fatty acid identified. On the basis of the fatty acid identified, the quality indices were calculated using the equations proposed by Hlbricht and Southgate (1991). Data were analysed by GLM procedure of SAS (2001).

III – Results and conclusions

Relative percentages of individual fatty acids in intramuscular neutral lipids fractions of *Psoas major* muscle and of two diets are shown in Tables 1 and 2.

Table 1. Fatty acid classes in two diets

Fatty acids	Acorn		Barley	
	Mean	±SD	Mean	±SD
∑SFA	20.76	0.06	30.31	0.53
∑MUFA	56.43	0.31	17.89	0.07
∑PUFA	22.81	0.37	51.80	0.61
∑PUFA n -3	2.87	0.05	7.18	0.28
∑PUFA n -6	19.93	0.32	44.62	0.33

Results are expressed as percentage of total fatty acid methyl esters identified; SFA, saturated fatty acids; MUFA, monounsaturated fatty acids; PUFA, polyunsaturated fatty acids).

Table 2. Fatty acid profiles (% methyl esters listed) of intramuscular fat of *Psoas major* muscle from Nero Siciliano pigs fed with different fattening diets

Fatty Acids	Feeding system				*P*-value
	Acorn		Barley		
	Mean	±SD	Mean	±SD	
C12	0.07	0.01	0.07	0.01	ns
C14	1.23	0.11	1.17	0.16	ns
C15	0.07	0.02	0.06	0.01	ns
C16	22.87	1.23	23.74	0.90	0.0001
C16:1	3.44	0.34	3.50	0.31	ns
C17	0.30	0.04	0.28	0.04	0.045
C17:1	0.29	0.17	0.29	0.11	ns
C18	13.23	3.08	13.66	1.01	ns
C18:1 n-9	39.58	3.38	43.06	1.48	0.011
C18:1 n-7	7.82	3.33	4.61	0.44	0.016
C18:2 n-6	7.11	1.03	6.19	0.84	0.001
C18:3 n-3	0.36	0.09	0.26	0.03	0.0002
C20	0.26	0.07	0.27	0.06	ns
C20:1 n-9	1.00	0.10	0.93	0.22	ns
C20:2 n-6	0.38	0.05	0.31	0.12	0.006
C20:3 n-6	0.09	0.04	0.11	0.09	ns
C20:4 n-6	1.30	0.65	0.98	0.20	0.005
C22	0.14	0.07	0.09	0.03	0.003
C22:1 n-9	0.08	0.07	0.06	0.01	ns
C22:5 n-3	0.15	0.08	0.18	0.06	ns
C24	0.14	0.03	0.09	0.06	0.0001
C22:6 n-3	0.09	0.04	0.09	0.04	ns
∑SFA	38.42	3.86	39.51	1.73	ns
∑MUFA	52.21	2.95	52.39	1.32	ns
∑PUFA	9.50	1.80	8.16	1.12	0.001
∑PUFA n -3	0.61	0.15	0.53	0.10	0.038
∑PUFA n -6	8.89	1.68	7.63	1.05	0.001
AI	0.45	0.05	0.47	0.03	ns
TI	1.16	0.21	1.22	0.10	ns

Results are expressed as percentage of total fatty acid methyl esters identified. AI, atherogenic index; TI, thrombogenic index; SFA, saturated fatty acids; MUFA, monounsaturated fatty acids; PUFA, polyunsaturated fatty acids).

The polyunsaturated fatty acids (PUFA) showed an increase (P=0.001) in group A compared with group B, accompanied by a decrease in the total saturated fatty acids (SFA). Particularly, the PUFA n-3 group was highest in muscle of pigs fed with acorn (0.61% *vs* 0.53%; P=0.038) as well as PUFA n-6 group (8.89% *vs* 7.63%; P=0.001). This difference is due mainly to the essential fatty acids (EFA) (Givens *et al*., 2006), present in greater proportion in the acorn group. The α-linolenic acid (C18:3 n-3) and the linoleic acid (C18:2 n-6), in fact, were 0.36% and 7.11% respectively in acorn group and 0.26% and 6.19% in barley group. Also the arachidonic acid (important product from EFA by the action of Δ5 and Δ6 desaturase and elongase enzymes), which has various metabolic roles including eicosanoid production (Wood *et al.*, 2008) is increased in pigs fed with acorn (1.30% *vs* 0.98%; P=0.005). The PUFA content in muscle from pig fed with barley (8.16%) is significantly (P=0.001) lower than in acorn group (9.50%); this results suggest that in A group, the fast-oxidative metabolism of *Psoas* muscle, was slowed by polyphenols, and/or some derived antioxidant metabolites, present in acorn.

In recent years there has been much interest in the beneficial effects of the very long chain (VLC, carbon chain length ≥20) PUFA, in particular eicosapentaenoic acid (EPA, C20:5) and docosahexaenoic acid (DHA, C22:6). The beneficial effects have been well documented and include anti-atherogenic, anti-thrombotic and anti-inflammatory effects. Crucially, it is now evident that in vivo synthesis of EPA and DHA from dietary α-linolenic acid (ALNA; C18:3 n-3) is very limited in adult humans, especially in men (Burdge *et al.*, 2003), so it is interesting to examine how increasing intake of ALNA in human diet through animal nutrition. A number of intervention studies do suggest that high intakes of ALNA can beneficially affect a number of CVD risk factors including LDL cholesterol (e.g. Zhao *et al*., 2004). The result for α-linolenic acid content in acorns analysed for this study (2.08%) was higher than previously reported (1.8%) by Petrović *et al*. (2004) and higher than amounts of C18:3 n-3 (0.7-1.0%) usually found in acorns of Mediterranean forest oaks (*Q. ilex, Q. Rotundifolia* and *Q. suber*) (Cava *et al*., 1997; López-Bote, 1998). Feeding acorn raised the n-3 content in the meat, being of interest from the consumer's health point of view. Additional researches however are needed .

Acknowledgements

The authors wish to acknowledge the financial support received by MIUR- PRIN 2005 and by PRA 2005 (Prof. A. Zumbo).

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Ripening technology and microbial biodiversity in the preparation of Nebrodi salame

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Abstract. The production of salame in the Nebrodi area, north-eastern Sicily, is carried out by craftmade technique. An important role in the transformation of the pork meat in salame is done by the technique of preparation and by microorganisms present. Next to a specific technology that provides a technique for preparation / maturation with different characteristics (type of casing, drying / ageing environments) is present a microbial biodiversity that is most evident in a surprising large distribution of species present at a wide range of different biotypes both in reference to lactic acid bacteria and staphylococci. The habitats considered, represented by different samples of salame produced in the Nebrodi area using mature Black Pig meat, show a substantial wholesomeness of meat products where the dominant microflora is represented by specific lactic acid bacteria (*Lactobacillus sake, Lactobacillus curvatus* and *Lactobacillus plantarum*) and stafilococci (*Staphylococcus xylosus* largely dominant, *Staphylococcus equorum* and other minority species). The microflora has shown a complete reliability with good organoleptic and healthiness characteristics of salame, and is available, if necessary, to integrate in a "natural" way the production technology of sausage produced in Nebrodi.

Keywords. Salami – Nebrodi – Ripening – Microorganism – Technology – Black pig.

Technologie de maturation et biodiversité microbienne dans la préparation du salami de Nebrodi

Résumé. *La production de salami dans le Nebrodi, au nord-est de la Sicile, est effectuée artisanalement. La technique de préparation et les microorganismes présents jouent un rôle important dans la transformation de la viande de porc en saucisson. À côté d'une technologie spécifique qui fournit une technique pour la préparation / maturation avec différentes caractéristiques (le type d'enveloppe, les environnements de séchage / maturation) il y a une biodiversité microbienne qui se manifeste surtout dans un nombre surprenant d'espèces présentes dans une large gamme de différents biotypes à la fois en termes de bactéries spécifiques d'acides lactiques et de staphylocoques. Les habitats considérés, représentés par différents échantillons de salami produit dans la zone de Nebrodi utilisant de la viande de porc noir, montrent une salubrité des produits dérivés, et des microflores représentées par des bactéries d'acide lactique (*Lactobacillus sake*,* Lactobacillus curvatus *et* Lactobacillus plantarum*) et des staphylocoques (*Staphylococcus xylosus *largement répandue,* Staphylococcus equorum *et d'autres en moindre nombre). Ces ferments ont montré une fiabilité complète avec de bonnes caractéristiques organoleptiques des produits, et sont disponibles pour les intégrer aux technologies de production de saucissons produits dans le Nebrodi.*

Mots-clés. *Salami – Nebrodi – Maturation – Caractéristiques microbiennes – Technologie – Porc noir.*

I – Introduction

In the processing of cured salami an important role is played by both the preparation technique and process parameters in the dough either by microorganisms that play a positive or negative role depending on their nature. The various microorganisms present in the meat can selectively degrade all components of meat lipids, proteins, sugars, etc., transforming them in countless elementary substances compared to those of beginning, not excluding toxic or malodorous. The control of microorganisms in meats, from raw materials is therefore of vital importance during seasoning.

Salami production in the Nebrodi area (Sicily) is generally carried out according to local traditions of craftmade technique. The preparation of the mixture is done to "a knife tip" using all the cuts of pork. Mixture is added salt, pepper whole and ground and natural essences. Stuffing is carried out in natural pork casing. The seasoning is conducted in natural environment, with an initial phase of drying, can be performed in air-conditioned rooms. Depending on the size of casing used and the environmental temperature and humidity conditions products with different ripening, between 30 and 120 days, are obtained. Genetic characteristics, hygiene and processing techniques maturation influence the final characteristics of the sausages (Diaferia *et al*.,2007, Pirone *et al*., 2007).

An important role on the control of final goods characteristics of the salame is done also by air temperature humidity used during the ripening process (Baldini *et al.,* 2000).

With present work, the thermohygrometric conditions and the microbiological biodiversity of Nebrodi salame have been studied.

II – Material and methods

1. Trend of relative humidity and temperature

They were recorded with the use of portable electronic recorders values of environmental temperature and relative humidity. The values recorded at intervals of 20 minutes were subsequently transferred and stored on personal computers. At intervals of 24 hours, mean, minimum and maximum were then calculated.

2. Isolation and identification of strains

Non-pathogenic strains of staphylococci were isolated and subjected to the following tests:

- biochemical profile by API STAPH (bioMérieux),
- oxidase (bioMérieux),
- coagulase (bioMérieux),
- lisostafine.

3. Technological characterization of isolates

The identified non-pathogenic strains of staphylococci were subjected to the following test technology :

- growth in presence of 5-10% NaCl at temperatures of 10-15-22°C,
- proteolytic activity in vitro,
- lipolytic activity,
- biogenic amines production,
- lisostafine resistance,
- lactic acid production.

4. Genetic analysis of strains of staphylococci and lactobacilli

The strains of staphylococci and lactobacilli considered for genetic testing have been reactivated by streaking on appropriate substrates, the colonies developed were removed and diluted in water to be lysed by microwaves .

DNA extracted from each strain was amplified by RAPD-PCR technique, which involves amplification of DNA fragments homologous to a bacterial specific primers and subsequent separation by electrophoresis on agarose gel.

The cycle of RAPD-PCR and primers used for identifying biotypes are shown in Table 1. For the identification of species belonging to the biotypes of lactic acid bacteria identified was run-16s rDNA sequencing, and for species identification of staphylococci belonging to the technique was applied ARDRA.

Table 1. RAPD-PCR and primers used for identifying biotypes

Nome primer	Sequence (5'-3')	Cycle
Rapd2	AgCAgCgTgg	Pre-hold: 94°C x 5min
		94°C x 1min
		29°C x 1min
		72°C x 2min
		20cycle
		from 29°C to 72°C time 90sec.
		94°C x 30sec
		55°C x 30sec
		72°C x 30sec
		45cycle
		Final-hold: 72°C x 5min

Conditions applied for PCR-RAPD.

III – Results and discussion

The identification with biochemical and molecular tests allowed the identification of 15 out of 16 strains as *S. xylosus*; strain (6BNE) was identified as *S. saprophyticus*. All strains grew at 10°C with 10% NaCl .

All rised at 15 and 21°C in 2 days even at pH 5.1, while at 10°C strains S16NE, S17NE, S18NE 8BNE and B did not grow in pH of 5.1.

At the latter pH value and temperature of 10°C strains 20NE and 18BNE rised in four days as all those tested at pH 5.4; others showed growth in 7 days.

The ability to grow at low temperature and low pH value is certainly significant from a technological standpoint.

Concerning the qualitative determination of proteolytic strains only 18BNE, S1NE, S18NE show a clear capacity to utilize protein.

Regarding qualitative tests for lipolityc activity only a strain showed metabolic activity toward fat throat while all strains caused tributyrin lipolysis, though with varying degrees of intensity. The quantitative test versus throat fat gave different results: all strains possessed metabolic capacity. The less active strain is 6BNE, the most active one is 8BNE B.

Regarding the production of lactic acid, the values found are obviously different depending on the substrate. In conditions similar to those of a mixture (NIM broth) produced amounts are not relevant (Iaccarino and Pirone, *2008)*.

A minimum of 0.083 g/l for the strain 6BNE and a maximum of 0.341 g/l for the strain 22BNE were found.

In media containing high levels of sugars (APT broth), behaviours are noticeably different than in NIM broth: the production of lactic acid is much higher in APT broth and the results much more variable among them.

Regarding the qualitative test for the production of biogenic amines, all strains are producers under adopted test conditions. The test is only indicative since it provides high concentrations of amino acid precursors.

Quantitative analysis of amino acids concentrations on the product at various stages of maturity, may give more complete and closer results to production conditions. In any case, the test run shows a strain (S18NE) less active because it decarboxylates only tryptophan.

The different production areas showed various thermo-hygrometric characteristics on the seasonality of manufacture (winter and spring), depending by use of natural environments (Table 2).

Table 2. Ranges of temperature and humidity recorded during the tests in different Nebrodi areas

Areas	Month	Temperature (°C)		Relative humidity (%)	
		min	max	min	max
Mirto(ME)	February	7.6	15.6	n.d.	n.d.
	March	7.8	13.5	n.d.	n.d.
	April	12.5	14.6	n.d.	n.d.
Piraino(ME)	February	10.5	17.1	43	86
	March	9.4	15.7	52	82
	April	12.7	14.7	70	75
Troina (EN)	February	10.4	19.6	57	74
	March	10.9	14.1	53	73
	April	12.6	17.6	44	63
Castiglione Siculo (CT)	February	12.1	15.1	45	80
	March	12.5	15.6	41	74
	April	13.3	14.3	55	84
Caronia(ME)	February	14.8	17.6	55	88
	March	15.1	17.5	62	78
	April	14.3	18.0	65	79
Longi(ME)	February	10.4	16.5	41	87
	March	9.9	15.8	45	80
	April	14.3	16.4	56	70
Sinagra(ME)	February	12.1	14.7	80	98
	March	11.6	15.5	53	98
	April	11.5	17.0	47	98

Regarding the genetic analysis, molecular fingerprinting to detect some biotypes of strains used, obtained by the technique of RAPD-PCR using the primer Rapd2, are shown in Figs 1 and 2, respectively for the staphylococci and lactobacilli .

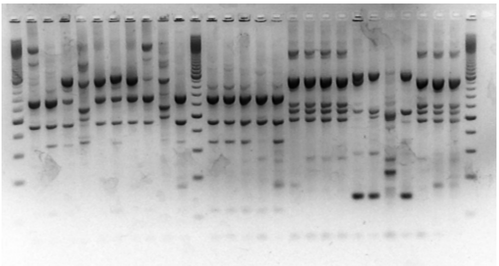

Fig. 1. RAPD profiles of staphylococci isolated from Nebrodi salame.

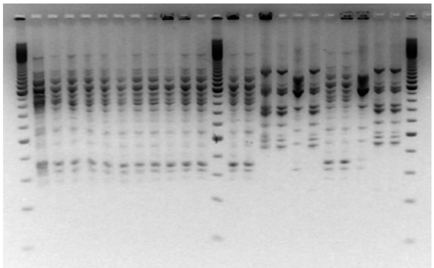

Fig. 2. RAPD profiles of lactobacilli isolated from Nebrodi salame.

5. Identification of species of lactic acid bacteria and staphylococci

A strain for each of identified biotypes of lactic acid bacteria were then subjected to sequence analysis of 16S-rDNA to identify the species 57 strains were identified.

To be able to identify the species of biotypes of stafilococci highlighted with the technique RAPD-PCR, 16S-rDNA of biotypes identified was instead analyzed by the genetic ARDRA technique; 129 strains of staphylococci were identified .

Table 2 and 3 indicate for the strains of lactobacilli and staphylococci, species determined using the ARDRA method, number of identified biotypes, number of strains.

Table 2 . Biotypes and species affiliation of strains of lactobacilli isolated

Biotypes belong to the species	Number of biotypes	Number of strains
Lactobacillus curvatus	3	9
Lactobacillus sakei	7	33
Lactobacillus plantarum	4	13
Waissella	1	2

Table3. Biotypes and species affiliation of strains of staphylococci isolated

Biotypes belong to the species	Number of biotypes	Number of strains
Staphylococcus equorum	4	37
Staphylococcus xylosus	12	59
Staphylococcus epidermidis	1	1
Staphylococcus saprophyticus	4	13
Enterococcus faecium	2	19

Acknowledgments

This research has been carried out within the framework of the project Animal Breeding: Quality Biodiversity Innovation Competitiveness . The publication is supported by the EC under the MED Programme . It reflects the author's view and the Community is not liable for any use that may be made of the information contained in this publication.

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Aroma components in Sobrasada of Mallorca from black pig

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Abstract. Sobrassada of Mallorca from black pig was used to determine the key aroma components. Sobrassada had a high content of monounsaturated fatty acids. A specific liberation of polyunsaturated free fatty acids (FFA) was detected in the proportion of FFA in contrast to the total fatty acid composition that was higher in polyunsaturated FA and, in lower proportion, monounsaturated ones. The analysis of the headspace of sobrassada resulted in the identification of eighty four different volatile compounds and 3 of them were for the first time detected in dry sausages (methyl nonanoate, 1-methyl-1H-pyrrole and 2-acetyl pyrrole). The aroma of sobrassada was characterized by thirty five different aroma active zones. These aroma active zones corresponded to compounds already detected as essential aroma contributors in dry sausages (3-methyl butanoic acid, ethyl 3-methyl butanoate, 2,3-butanedione, and acetic acid) and in addition compounds such as ethyl octanoate, furfural, benzaldehyde, (Z)-2-nonenal, 4-methyl-phenol, delta-hexalactone, heptanoic acid, 2-pentylfuran and 2-acetyl-pyrrole gave specific aroma notes.

Keywords. Sobrassada – Dry fermented sausage – Aroma – Volatile – Fatty acids.

Composants de l'arôme dans la Sobrassada de Majorque à partir de porc noir

Résumé. *La Sobrassada de Majorque à partir de porc noir a été utilisée pour déterminer les composants aromatiques clés. La Sobrassada avait une teneur élevée en acides gras monoinsaturés. Une libération spécifique d'acides gras polyinsaturés libres (FFA) a été détectée dans la proportion des FFA, contrairement à la composition en acides gras totaux qui a été plus élevée en gras polyinsaturés FFA et, en moindre proportion, en monoinsaturés. L'analyse de l´espace libre pour la Sobrassada a abouti à l´identification de quatre-vingt-quatre différents composés volatils, dont 3 ont été pour la première fois détectés dans les saucissons secs (nonanoate de méthyle, 1-méthyl-1H-pyrrole et 2-acétyl pyrrole). L'arôme de la Sobrassada a été caractérisé par trente-cinq arômes différents. Ces arômes correspondent à des composés déjà détectés en tant que contributeurs essentiels à l'arôme des saucissons secs (3-méthyl butanoïque, éthyle butanoate 3-méthyl, 2,3-butanedione, et acide acétique) et des additifs tels que octanoate d'éthyle, furfural, benzaldéhyde, (Z)-2-nonenal, 4-méthyl-phénol, delta-hexalactone, acide heptanoïque, 2-pentylfuran et 2-acétyl-pyrrole ont donné des notes aromatiques spécifiques*

Mots-clés. *Sobrassada – Saucisson sec – Arôme – Volatils – Acides gras.*

I – Introduction

Sobrassada is a dry cured meat product from the island of Mallorca (Spain) characterised by a high percentage of white fat (40-70%) together with lean pork meat (30-60%), additives such as curing agents, salt, nitrate and nitrite, and spices (paprika, pepper, origanum, etc.). The process consist on grinding the raw materials to obtain a fine paste that its filled into casing and left to ripen for several weeks to develop the typical sensory characteristics. Sobrassada of Mallorca from black pig is defined as a sobrassada exclusively processed using meat from Mallorca black pig and filled into natural casings. The lipid composition of the meat product as well as the lipolysis process affect its final flavour that in the case of Sobrassada of Mallorca from black pig could be essential to explain its characteristic aroma.

The highest aroma quality of traditional foods with protected geographical indication (PGI) meets consumer demands for less processed foods. The knowledge of those compounds responsible for the aroma in sobrassada is important to optimise the processing. Therefore our objective was to determine the key aroma compounds in Sobrassada of Mallorca from black pig and to study the contribution of lipolysis to the generation of the key aroma compounds.

II – Materials and methods

1. Sobrassada samples

Traditional dry cured sausages "Sobrassada of Mallorca from black pig", PGI (El Zagal, Felanitx, Mallorca, Spain) were used as described Gianelli *et al*. (2010).

2. Chemical analyses

The chemical parameters, pH, water activity (Aw), moisture and nitrogen content were determined as described Gianelli *et al*. (2010). Total lipids were extracted by the Folch method (Folch *et al*., 1957). The chemical analyses of each sobrassada sample were done in triplicate and results expressed as the mean in dry matter. Fatty acid methyl esters (FAME) of total lipids and free fatty acid analyses are described in Gianelli *et al*. (2010).

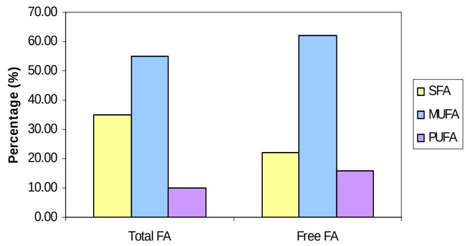
Extraction of headspace volatile compounds was done using a solid phase microextraction (SPME) device as described Gianelli *et al*. (2010). The extraction was done using a 85 µm CAR/PDMS stableflex fibre. For the identification and quantification of the volatile compounds, a gas chromatograph HP 7890A equipped with an HP 5975C mass selective detector (Hewlett Packard, Palo Alto, CA) was used. The compounds were separated on a DB-624 capillary column J & W Scientific (Agilent Technologies, USA) and analyzed as described Marco *et al*. (2007).

The gas chromatrography-olfactometry was done by analyzing the compounds adsorbed by the SPME fibre. The desorption of volatile compounds was done in a gas chromatograph (Agilent 6890, USA) with a FID detector and a sniffing port (split 2:1, respectively) (ODP3, Gerstel, Mülheim an der Ruhr, Germany) as described Gianelli *et al*. (2010). The detection frequency method was used to estimate the aromatic impact of each volatile compound. Three trained assessors evaluated the odors from the GC-effluent. Aroma compounds were identified by three different ways; comparison with mass spectra; comparison with the retention times of authentic standards injected in the GC-FID; and by coincidence of the assessors descriptors with those in the *Fenaroli's handbook of flavour ingredients.*

III – Results and discussion

1. Sobrassada composition

Sobrassada of Mallorca from black pig have a protein and fat contents of 15.3 % and 57.6 % respectively. The pH value was 4.6 and Aw was 0.81 which are in accordance with the values recommended by the PGI. Total fatty acid composition in Sobrassada of Mallorca from black pig is shown in Fig. 1. Total saturated fatty acids were about 34-35 %, monounsaturated 54-55 % and polyunsaturated 10 %. The free fatty acids (FFA) detected in Sobrassada had different proportions than the obtained for the total fatty acid concentration (Fig. 1). The proportion of FFA in contrast to the total fatty acid composition was higher in polyunsaturated FFA and, in lower proportion, monounsaturated ones.



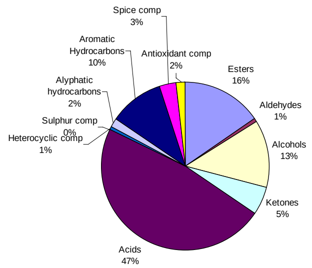


Fig 1. Total fatty acid and free fatty acid composition of sobrassada.

Fig. 2. Volatile compounds by chemical classes extracted from sobrassada using SPME.

2. Aroma analysis

The extraction of volatile compounds from the headspace of Sobrassada of Mallorca from black pig using SPME indicated the presence of high number of volatile compounds (Fig. 2).

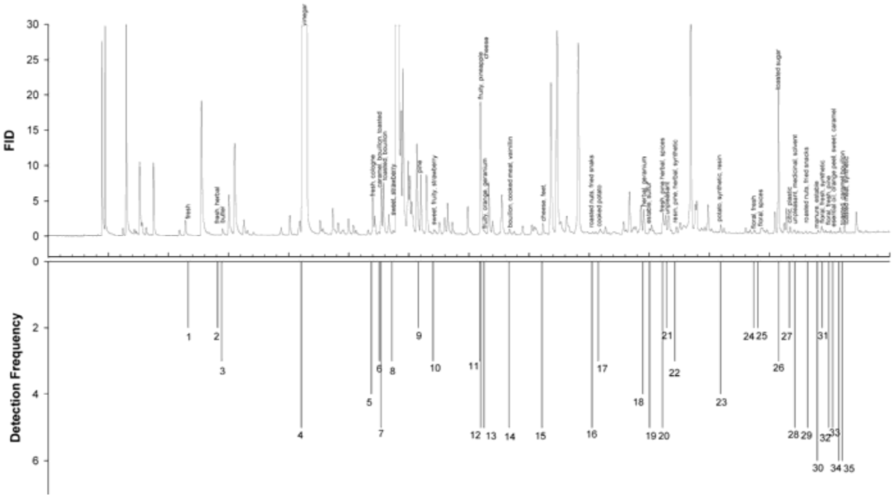


Fig. 3. Chromatogram and olfactogram of sobrassada from Mallorca of black pig.

The compounds identified corresponded to different chemical classes that represented a percentage of the total extracted area (Fig. 2). Thirty five different aroma active zones were detected (Fig. 3, Table 1), but 5 of them were not identified. Many of these compounds were also detected as aroma active compounds in dry sausages (Marco *et al*., 2007). The contribution of the compounds to the aroma of sobrassada can be evaluated by their detection frequency values (DF in Fig. 3). The compounds that showed the highest DF values in sobrassada were ethyl 3-methylbutanoate, ethyl octanoate, furfural, benzaldehyde, (Z)-2-nonenal, 4-methyl-phenol, delta-hexalactone, acetic, 3-methyl-butanoic and heptanoic acids, 2-pentylfuran, and 2-acetyl-pyrrole. In addition, 5 compounds contribute to the aroma with meaty notes such as ethyl octanoate, furfural, (Z)-2-nonenal, dimethyldisulfide, and 1-methyl-1H-pyrrole.

Table 1. Aroma active zones detected in sobrassada of Mallorca from black pig and chemical compound identified

N†	LRI [b]	Chemical Compound	GCO Descriptor	N†	LRI††	Chemical Compound	GCO Descriptor
1	602	2-methyl-propanal	fresh	19	1008	2-pentyl-furan	stable, sulfur
2	628	1-propanol	fresh, herbal	20	1021	benzaldehyde	fresh, pine, herbal, spices
3	631	2, 3 -butanodione	butter	21	1025	3-carene	unpleasant
4	701	Acetic acid	vinegar	22	1032	6-methyl-5-hepten-2-one	resin, pine, herbal, synthetic
5	766	Unknown	fresh, cologne	23	1076	hexanoic acid + alfa-terpinene	potato, synthetic, resin
6	773	dimethyl disulfide	caramel, Bouillon	24	1110	benzeneacetadehyde	floral, fresh
7	777	1-methyl-1H-pyrrole	toasted, Bouillon	25	1115	trans-2-octenal	floral, spices
8	785	Unknown	sweet, strawberry	26	1141	tetramethyl-pyrazine	toasted sugar
9	810	1-pentanol	pine	27	1155	nonanal	citric, plastic
10	825	ethyl butyrate	sweet, fruity, stawberry	28	1162	heptanoic acid	medicinal, solvent, rancid
11	871	ethyl 2-methyl-butanoate	fruit, pineapple	29	1178	2-acetyl pyrrol	roasted nuts, fried snacks
12	872	Unknown	cheese	30	1190	4-methyl-phenol	manure, stable
13	875	ethyl 3-methylbutanoate	fruity, orange, geranium	31	1196	phenylethyl alcohol	floral, fresh, synthetic
14	900	furfural	bouillon, cooked meat	32	1206	Unknown	floral, fresh, pine
15	925	3-methyl-butanoic acid	cheese, feet	33	1213	delta-hexalactona	essential oil, orange peel
16	963	Unknown	roasted nuts, snacks	34	1223	(Z)- 2-nonenal	toasted caramel bouillon
17	968	3- (methylthio)-propanal	cooked potato	35	1229	ethyl octanoate	toasted meat synthetic.
18	1002	beta-myrcene	herbal, geranium				

†Number of the aroma active zones as represented in Fig. 3.
††Linear retention indices of the aroma detected in the sniffer port.

IV – Conclusions

The aroma of sobrassada of Mallorca from black pig was not only due to compounds already reported as essential contributors to the aroma of dry sausages (3-methyl butanoic acid, ethyl 3-methyl butanoate, 2,3-butanedione, and acetic acid) but also to the presence of other compounds such as ethyl octanoate, furfural, benzaldehyde, (Z)-2-nonenal, 4-methyl-phenol, delta-hexalactone, heptanoic acid, 2-pentylfuran and 2-acetyl-pyrrole which gave specific aroma notes. Many of these compounds are derived from the lipid autooxidation process.

Acknowledgments

Financial support from AGL 2009-08787 from MCINN (Spain) and FEDER funds and FONDECYT for the 11070128 (Chile) are fully acknowledged. The predoctoral schorlarship from GVA (Generalitat Valenciana, Spain) to A. Olivares is also acknowledged.

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Effect of sex and slaughter weight on carcass traits measured *in vivo* with ultrasound and *post-mortem* in the carcass in Iberian pigs

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Abstract. Carcass composition of Iberian pigs is very important because it influences the quality of their cured products. Therefore, the objective of this study was to determine the effects of sex and slaughter weight on carcass traits measured *in vivo* with ultrasound and subsequently in the carcass. One hundred and sixty five castrate males and females with a weight ranging from 130 to 175 kg were ultrasound-scanned before slaughter with a 12-cm long, 3,5 Mhz probe. Two images were collected across the *Longissimus dorsi* muscle, one between the 10th and 11th ribs and another after the last rib. After slaughtering, carcass, hams, shoulders and loins were weighted, and the same measurements performed for the ultrasound images were also done in the carcass as follows: loin area, and fat thickness (external, middle, internal, external + middle layers and total). Sex effects were significant for shoulder traits, for ultrasound fat thickness measurements taken between 10th and 11th ribs, and also for some other traits at the last rib level. Slaughtering pigs over 165 kg did not improve carcass or cut yields.

Keywords. Iberian pig – Carcass traits – Backfat – Slaughter weight – Ultrasound.

Effet du sexe et du poids à l'abattage sur des caractères de la carcasse mesurés en vif par ultrasons et sur la carcasse post mortem pour le porc Ibérique

Résumé. *Dû à l'importance que les caractères de composition de la carcasse du porc ibérique ont sur la qualité des produits séchés, on a étudié l'effet du sexe et du poids à l'abattage sur ces caractères, qui avaient été mesurés avant l'abattage par écographie, et ultérieurement sur la carcasse. On a abattu 165 mâles et femelles castrés, avec un poids entre 130 et 175 kg. Avant l'abattage ils ont été échographiés avec une sonde de 3,5 Mhz et 12 cm. On a récupéré deux images échographiques perpendiculaires au* Longissimus dorsi*, l'une entre les côtes 10 et 11, et l'autre après la dernière côte. Après l'abattage on a pesé la carcasse, les jambons et les épaules, et les filets, et on a réalisé sur la carcasse les mêmes mesures qu'avec les images échographiques : surface du filet et épaisseur totale et partielle de la graisse dorsale dans les couches extérieure, moyenne, interne, externe + moyenne et total. Le sexe a affecté les caractères des épaules, les épaisseurs ultrasons-graphiques mesurées entre les côtes 10 et 11, et diverses épaisseurs de la dernière côte. L'abattage d'animaux ayant plus de 165 kg n'a pas signifié une amélioration des rendements de la carcasse ou des coupes.*

Mots clés. *Porc Ibérique – Caractères de la carcasse – Graisse dorsale – Poids d'abattage – Ultrason.*

I – Introduction

Fat tissue deposition is very important for Iberian pig production, since the quality of Iberian pig meat depends on the amount and distribution of fat. Iberian pigs are finished in two well differentiated systems: one is based in the use of concentrates, with the animals being in general a crossbred between Iberian females and Duroc males that are raised either indoors or outdoors are slaughtered at around ten months of age. The other system is based in the use of natural resources (acorns and grass) and the animals are pure breed Iberian pigs, which are raised outdoors and slaughtered at 17-18 months of age. This second system is the most traditional and yields a high quality meat (with a high percentage of intramuscular fat) which is very appreciated by consumers. However, to reach this high percentage of intramuscular fat, a large amount of subcutaneous back fat cover (around 90 mm thick) and large deposits of internal fat must be accumulated. This back fat thickness is divided into three layers that behave differently depending on the age or breed of the pigs (Eggert and Schinckel, 1998; Alfonso 2004). Thus, the aim of this study is to determine differences in fat layers' thickness due to sex and slaughter age as measured at two different rib locations *in vivo* with ultrasound and *postmortem* in the carcass in Iberian pigs.

II – Materials and methods

Animals: For this study, 163 Iberian pigs of the red *("Retinto"*) strain were used. Animals were raised indoors in standard commercial conditions up to 25 kg of body weight (BW). After this period, animals were raised outdoors in extensive conditions and fed a concentrate in a restricted diet (as usual in this traditional system) up to 12 months of age. At this age, animals were divided in five groups and started an "ad libitum" feeding period with different types of food: One group was fed only pasture and acorn; the second was fed pasture and acorn up to 135 kg of BW and then finished with a standard, commercial concentrate; the third group was fed pastures, acorn and standard concentrate at the same time; the forth group was fed standard concentrate; and the fifth group was fed a commercial concentrate having a high level of oleic acid to imitate acorn composition. Animals were fed for a period of 2.5 to 3 months and were slaughtered after reaching 135 kg of BW (135 to 175 kg range). To allow for a thorough study of carcasses, no more than 20 animals at a time were slaughtered.

Ultrasound image collection: One day before slaughter, pigs were ultrasonically scanned to measure body composition, using an Aloka 500 apparatus (Aloka Holding - Europe, Switzerland) and a 3,5 MHz, 12 cm long probe. Ultrasound images were collected by placing the probe perpendicular to the loin a two different rib levels: one image was taken between the 10^{th} and 11^{th} ribs (10^{th} intercostal space), and the other was taken just behind the last (14^{th}) rib. A soft, rubbery adaptor made of Supperflap® was used between the animal and the probe to allow for an adequate contact despite of the curved back surface. Images were digitalized and stored in a computer. Image measurements were done afterwards by using the Biosoft® software (Biotronics inc., Ames, IA, USA). For each image, loin area and fat thickness of the 3 layers were measured. The ultrasound ("U") measurements, taken at the 10^{th} intercostal space and behind the last rib, respectively ("10", "14"), were the following: external ("E") back fat layer (F10EU and F14EU); middle ("M") layer (F10MU and FG14MU); internal ("I") layer (F10IU and F14IC); external + middle ("EM") layers (F10EMU and F14EMU); and total ("T") backfat thickness (F10TU and F14TU).

Carcass data: After slaughter, carcass weight and weight of commercials cuts (ham, shoulder and loin) were measured. In addition, a portion of loin containing 4 chops (spanning from the $11^{\mathbf{th}}$ to the$14^{\mathbf{th}}$ ribs) was extracted from the carcass and used to measure back fat layers thickness and loin area at the two already mentioned rib levels.

Data was analyzed by using GLM (General Linear Models) procedures in SAS.

III – Results and discussion

Sex effects were not significant for the thickness of any of the back fat layers measured in the carcass at the 10^{th} intercostal space, as shown in Fig. 1. However, sex had a significant effect on the external and middle layer thickness when measured with ultrasound, so that castrate females had more back fat thickness than castrate males. Latorre (2004) indicated that castrate males had more backfat than intact females. Measurements taken at the last rib level were significant for the middle (measured with ultrasound) and the internal layer (measured with both techniques), but not for the external one, as shown in Fig. 2. Total back fat was significantly different for sex effects at the 10-11 rib only when measured with ultrasound, and at the 14 rib, either measured in the carcass or with ultrasound. Back fat was thicker at the 10-11 rib level than behind the last rib, and this difference was due to differences in the middle layer. In relation to slaughter weight effects, there were no significant differences at the 10th intercostal space in external and internal fat layers once animals reached 155 kg of weight. However, there were significant differences between animals lighter than 155 kg and the heaviest ones, as shown in Fig. 3.

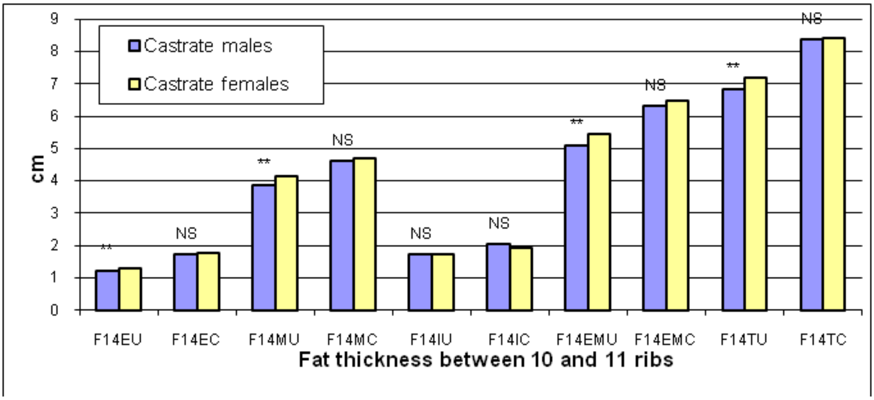

Fig. 1. **Effect of sex on the 3 backfat layer thicknesses (E:external; M:middle; I:internal; EM:E+M; T:total) measured with ultrasound (U) and in the carcass (C) at the 10th intercostal space.**

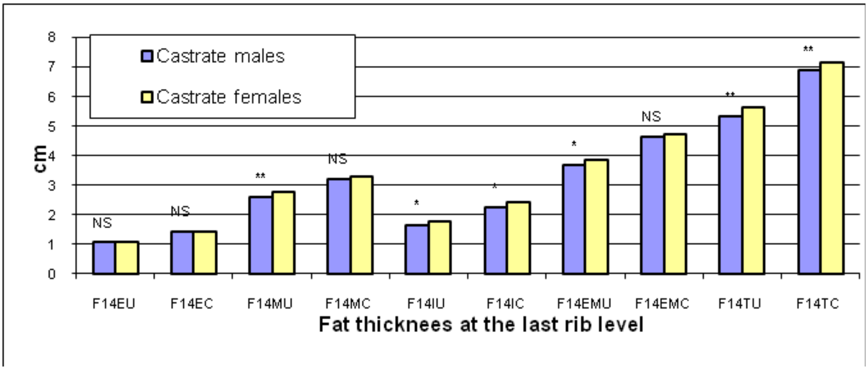

Fig. 2. **Effect of sex on the 3 backfat layer thicknesses (E:external; M:middle; I:internal; EM:E+M; T:total) measured with ultrasound (U) and in the carcass (C) behind the last rib (14).**

This indicated that the growth of these layers is slow after this weight, with no significant differences between the 155-165 kg and 166-175 kg classes. This results are also similar for total backfat. However, different results were found by Lo Fiego *et al*. (2005), because these authors concluded that pigs with 175 kg on average have thicker total backfat than those averaging 151 and 164 kg. With respect to the middle layer, there were significant differences among the three weight classes if measured with ultrasound, suggesting that this layer has a greater metabolic activity, as stated earlier by Leymaster and Mersmann (1991). Alfonso (2004) also indicated that the middle layer explained more than 50% of the overall back fat differences between two breeds. Results at the 14 rib level (Fig. 4) were similar to results at the 10-11 rib, with the exception that there were no differences among the three weight classes for the middle layer.

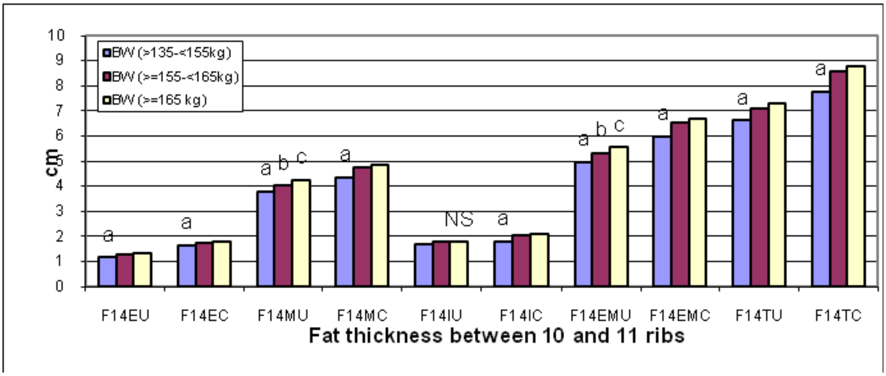

Fig. 3. Effect of slaughter weight on backfat layers (E:external; M:middle; I:internal; EM:E+M; T:total) measured with ultrasound (U) and in the carcass (C) at the 10th intercostal space.

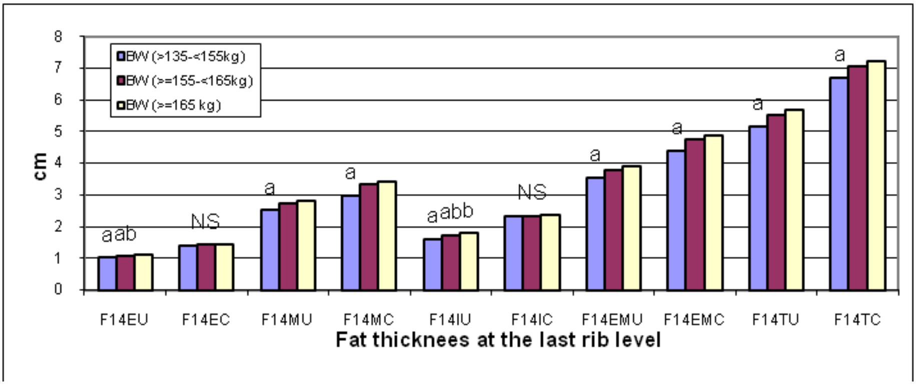

Fig. 4. Effect of slaughter weight on backfat layers (E:external; M:middle; I:internal; EM:E+M; T:total) measured with ultrasound (U) and in the carcass (C) behind the last rib (14).

IV – Conclusions

Backfat layers do not follow an uniform growth pattern. Middle backfat layer growth suggests a greater metabolic activity than the other two layers. Backfat is thicker at the 10-11 rib level than at the last rib level.

Acknowledgements

The authors want to thank FEDER, (PDT09B039) , INIA (RTA2007-0093-00-00) and Junta de Extremadura for financial support.

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Presentation's effect of granulated or wet barley during the finishing phase on the productive yield of Majorcan Black Pig

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Abstract. Majorcan Black Pig is an autochthonous breed in danger of extinction which is preserved due to its use to elaborate Sobrassada de Mallorca. Sustainability of this production system depends on the economic margin which is generated, thus the use of raw materials avoiding industrial processing is of a great importance. 20 castrated males were distributed in two batches, each one in a 1.5 ha plot placed contiguous, with an initial weight of 105 kg and during 86 days. The experimental design considered as the fixed effect the way the feed was presented: barley that was moistened in a water container during 24 hours *vs* dry granulated barley. In both cases was used the same barley origin and composition. Animals fed by dry granulated barley showed higher growth rates than those fed by moistened barley (543 *vs* 471 g/day, p<0.05) and higher feed intakes (2.76 *vs* 2.55 kg/day, p<0.05). Despite of these results regarding productive efficiency, the economic benefit generated by feeding the animals with moistened barley was 34 € per finishing pig.

Keywords. Majorcan Black Pig – Sustainability – Moistened barley.

Effet de l'alimentation au cours de la phase de finition avec de l'orge humide ou de l'orge granulée sur le rendement de production du porc noir de Majorque

Resumé. *Le porc noir de Majorque est une race autochtone menacée d'extinction qui est utilisée dans la préparation de la Sobrassada de Majorque. La durabilité de leur production dépend de la marge économique obtenue pour la viande et la graisse de ces animaux. À cet égard, et afin de réduire l'impact économique de l'alimentation animale, il est essentiel d'utiliser des matières premières sans transformation. Nous avons utilisé un total de 20 mâles castrés, divisé entre deux parcelles adjacentes similaires et de 1,5 ha dans la zone, avec un poids de départ de 105 kg dans un essai de 86 jours. Le traitement expérimental a été mené dans un seul but, la présentation de l'orge (hydratée pendant 24 h ou granuleuse et sèche). L'origine et la composition de l'orge étaient les mêmes dans les deux traitements. Les animaux nourris avec des granulés secs d'orge ont montré une plus forte croissance (543 g/j* vs *471, P <0,05) et consommé plus de nourriture (2,76 kg/j* vs *2,55 P <0,05). Il n'y avait pas de différences significatives concernant les taux de conversion. En dépit de ces résultats techniques, en termes économiques, la marge brute par porc générée par le régime avec de l'orge trempée était supérieure de 34 € à la marge obtenue par l'orge granulée.*

Mots-clés. *Porc noir de Majorque – Durabilité – Orge humide.*

I – Introduction

The Majorcan black pig is a breed in danger of extinction and is preserved by its use in developing Sobrassada de Mallorca de Porc Negre is profitable for the farmer (Jaume *et al.,* 2006) . Its production is extensive and is characterized by the use of endogenous resources in their diet, both as feeder grazing. The grass is supplemented with cereals and legumes. Cereals are usually subjected to a grinding treatment, transformed into flour. This process has an economic cost that is offset by the increased palatability and digestibility of the meal in front of whole grains (Valencia *et al*., 2008). Milling cost is not insignificant and can make about 10 cents per kg. Sustainability of this production system depends on the economic margin which is generated, thus, the use of raw materials without processing industry, such as wet barley, is of great importance. The aim of this work was to study presentation's effect granulated or wet barley during the finishing phase on the productive yield of Majorcan Black Pig.

II – Materials and methods

Diets are composed of barley and peas in the proportions (g/100 g; fresh matter basis) of 80:20. The dietary treatments consisted of two presentations of barley granulated (M) and wet for a day (B). They have established two experimental groups with 10 barrows Majorcan Black Pig bread each. At the start of the test the average weight of animals was 105 kg and they were distributed in two batches, each one in a 1.5 ha plot placed contiguous during 86 days. The experimental design considered as the fixed effect the way the feed was presented: barley that was moistened in a water container during 24 hours vs. dry granulated barley. In both cases was used the same barley origin and composition. The animals were weighed individually at the beginning (Pi) and at the end of the test (Pf) and food supplied daily to each group (Co). We used student t-test statistical package Stratgraphics to analyze whether the differences between batches are significant. It takes $p < 0.05$ as a minimum level of significance.

III – Results and discussion

All the pigs remained in good health throughout the experimental period. The presentation's effect are presented in Table 1. During the finishing period (86 days) animals fed by dry granulated barley grew faster than those fed by moistened barley, so the average daily gain (GMD) in group M was 543 g/day and in group 471 g/day. Feed intake was significantly higher in group M than in B (2.76 *vs* 2.55 kg/day). Consequently, feed conversion ratio (IC) of group M was significantly better (5.21) than those observed in pigs of group M (5.54).

Table 1. Effect of the presentation of barley in growth performances

Treatment	N	Pi (kg)	Pf (kg)	GMD (g/day)	IC
M	10	105	151.57^{a}	530.34^{a}	5.21^{a}
B	10	105	145.78^{b}	460.00^{b}	5.54^{b}

a,b Different superscripts across rows indicate significant differences.

It has been estimated gross margin per animal using the technical data of Table 1, and the prices of whole barley (0.130 €/kg) and barley granulated (0.228 €/kg). So it turns out, that gross margin is 34 € more in animals fed wet barley.

IV – Conclusions

Under the conditions of the present experiment, the use of wet barley has not produced better results in the final growth of the Majorcan Black Pig that barley granulated. Yet economic performance has shown the opposite trend and the gross margin is higher in the case of barley wet. This aspect is very important from the standpoint of sustainability of Majorcan Black Pig.

Acknowledgements

The authors want to thank the pig producers from Sa Cova Nova in Manacor, Mallorca, for their contribution in the present study. The research was supported by the RAPE 2009 grants.

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Effect of packaging material on volatile organic compounds (VOCs) of sliced and MAP packaged typical Italian and Spanish dry-cured hams

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Abstract. The volatile organic compounds (VOCs) of Parma and Teruel dry-cured hams were studied, by comparing fresh and MAP (modified atmosphere packaging) sliced products. Two polymeric food-grade packaging materials were used for ready-to-eat packages of pre-sliced dry-cured hams: PET (polyethylene terephthalate) with barrier film PET/EVOH/PE (polyethylene terephthalate / ethylene-vinyl alcohol / polyethylene copolymer) and PLA (polylactic acid) with PLA barrier film. Several differences in VOCs were attributed to dry-cured ham nature, with ethyl esters being a main feature of Parma ham, while compounds generated by amino acids catabolism prevailed in Teruel ham. Packaging in PLA increased VOCs originated from lipid oxidation, more abundant in Parma hams, and from amino acid catabolism, while packaging in PET increased the signals due to branched-chain alkanes (BCAs). VOCs originated from carbohydrate fermentation were negligibly affected by packaging material, while a dependence from the nature of dry-cured ham was observed.

Keywords. Dry-cured ham – Packaging material – Volatile organic compounds – Polylactic acid.

Effet du matériel de conditionnement sur les composés organiques volatils (COV) des jambons italiens et espagnols en tranches et conditionnés en atmosphère modifiée

Résumé. *Les propriétés du jambon de Parme et du jambon de Teruel en termes de composés organiques volatils (COV) ont été comparées, par comparaison entre les produits frais en tranches et les produits emballés en atmosphère modifiée. Pour le conditionnement du jambon prétranché on a utilisé deux matériaux d'emballage de qualité alimentaire, consistant en deux polymères différents: PET standard (polyéthylène téréphtalate) avec barrière PET / EVOH / PE (polyéthylène téréphtalate / éthylène-alcool vinylique / copolymère de polyéthylène) et PLA (acide polylactique) avec des films barrière en PLA. Plusieurs des différences de COV observées sont attribuées à la nature du jambon, notamment les esters éthyliques qui caractérisent le jambon de Parme, et au contraire, les composés produits par le catabolisme des acides aminés ont plus d'impact dans le Jambon de Teruel. L'emballage en PLA a montré une augmentation des COV originaires de l'oxydation des lipides et du catabolisme des acides aminés, alors que l'emballage en PET a causé une augmentation de l'intensité d'alcanes à chaîne ramifiée (BCAs). Les COV provenant de la fermentation des glucides ont été négligeablement affectés par le matériel d'emballage, tandis que leur dépendance par rapport à la nature du jambon a été démontrée.*

Mots-clés. *Jambon – Matériaux d'emballage – Composés organiques volatils – Acide polylactique.*

I – Introduction

Protective atmosphere packaged (MAP) pre-sliced dry-cured hams gained in recent years a positive commercial trend (+7.8% in 2009, source: Consortium of Parma ham, 2010). Recent studies (Parolari *et al*., 2009) reported the occasional onset of changes in MAP packaged dry-cured hams, impairing colour, odour and taste. The standard packaging material used for dry-cured ham is the food-grade polyethylene terephthalate (PET). Meanwhile, new materials have been investigated, taking into account environmental sustainability too. Among them, the biopolymer polylactic acid (PLA) has grabbed attention because it is synthesized from processed corn and it biodegrades after use. PLA is regarded like a "natural" package with good flavor retention; a limiting factor for PLA is its relatively poor barrier to water vapor and O_2 (Rhim *et al*., 2009). In this study, the VOCs were investigated, to focus differences due to ham nature and packaging.

II – Materials and methods

Four Parma (P) and 4 Teruel (T) dry-cured hams, aged 17-20 months, were sliced and packaged in PLA and PET with N_2:CO_2 = 70:30 MAP. Each package was filled with 85-90 g of 15 mm-thick slices. VOCs analysis was carried out on fresh and packaged slices (starting 2 weeks after packaging): 3 g, finely cut with a knife, were subjected to HS-SPME-GC-MS with a CAR/PDMS/DVB fiber (Supelco) for 120 min at 40°C, according to the method of Pinna *et al*. (2009). Data were checked for normal distribution and analyzed by the General Linear Model (GLM) procedure of SPSS ver. 11.5. The calculated model included ham nature and packaging as main effects. The Least Square Means (LSM) were estimated and the Bonferroni t-test was performed to statistically separate them. Principal Component Analysis (PCA) was run and scores of ham samples were graphically plotted onto the PC1-PC2 plane.

III – Results and discussion

Aroma compounds are reported in Table 1 and grouped according to origin mechanism.

Table 1. Effect of ham nature and packaging type on VOCs (expressed in AU × 10^{-4})

Volatile compounds†	Ham nature (N)		Packaging (P)			Significance			Loadings††	
	Parma	Teruel	None	PET	PLA	N	P	O x P	PC1	PC2
Lipid Oxidation										
1-Pentanol	95.8^{a}	46.9^{b}	54.6^{b}	54.8^{b}	105^{a}	***	***	ns	0.63	0.04
Pentanal	72.4^{a}	30.6^{b}	51.2	49.2	54.3	***	ns	ns	0.71	0.04
Hexane	9.70	5.46	7.94	7.76	7.14	*	ns	ns		
1-Hexanol	152	88.6	69.5	119	172	*	*	ns	0.52	0.29
Hexanal	440^{a}	143^{b}	225	248	401	***	*	ns	0.79	0.16
Heptane	27.6^{a}	15.1^{b}	17.1	17.8	29.2	**	ns	ns	0.66	0.05
Heptanal	152^{a}	63.8^{b}	89.7	98.7	136	***	ns	ns	0.79	0.17
2-Heptenal	7.23^{a}	2.82^{b}	4.69	2.99	7.39	**	ns	ns		
3-Heptanone	173	136	97.6^{b}	116^{b}	250^{a}	ns	***	ns		
Heptanoic Acid	268^{a}	103^{b}	139^{b}	159^{ab}	254^{a}	***	**	ns	0.86	0.19
Octane	184.3^{a}	97.1^{b}	100.4	119.6	201.9	**	*	ns	0.66	0.14
Octanal	54.3^{a}	26.5^{b}	41.1	39.2	41.0	***	ns	ns	0.70	0.11
1-Octen-3-ol	54.1^{a}	29.9^{b}	17.8^{b}	30.3^{b}	78.0^{a}	**	***	ns	0.59	0.11
Nonane	5.19^{a}	3.17^{a}	2.88^{b}	3.48^{b}	6.20^{a}	**	***	ns		
2-Nonanone	12.9	9.51	9.53	8.32	15.8	*	ns	ns	0.50	-0.06
2-Decenal	12.2^{a}	4.1^{a}	4.49^{b}	8.53^{ab}	11.4^{a}	***	***	*	-0.38	0.76
Undecane	18.5	24.5	12.1^{b}	33.9^{a}	18.5^{ab}	ns	***	ns	0.63	0.04
Carbohydrate fermentation										
Ethanol	679^{a}	317^{b}	410	485	762	***	ns	ns	0.62	0.28
Acetic Acid	78.2	63.8	63.6	57.8	91.6	ns	*	ns	0.62	-0.08
2-Butanone	40.3^{b}	77.0^{a}	66.0	60.1	49.9	***	ns	ns	-0.64	-0.29
2,3-butanedione	3.27^{b}	4.67^{a}	3.63	3.71	4.56	**	ns	ns		
3-hydroxy-2-butanone	99.1^{b}	165.9^{a}	152.4	138.1	107.0	**	ns	ns	-0.57	-0.03
Butanoic Acid	163^{a}	104^{b}	132	117	151	***	ns	ns	0.71	-0.07

Table 1. (*cont.*) Effect of ham nature and packaging type on VOCs (expressed in AU × 10^{-4})

Volatile compounds†	Ham nature (N)		Packaging (P)			Significance			Loadings††	
	Parma	Teruel	None	PET	PLA	N	P	O x P	PC1	PC2
Amino acid catabolism										
2-Methyl-propanal	13.1^{b}	24.7^{a}	16.5^{b}	18.1^{ab}	22.2^{a}	***	**	ns	-0.61	-0.19
3-Methyl-thio-propanal	10.5^{b}	17.4^{a}	9.54^{b}	10.5^{b}	21.8^{a}	**	***	**		
2-Metihyl-1-butanol	32.8	35.3	21.9^{b}	29.5^{ab}	50.7^{a}	ns	**	ns		
2-Methyl-butanal	127^{b}	254^{a}	161	209	201	***	ns	ns	-0.67	-0.05
3-Metihy-butanal	189^{b}	267^{a}	205	223	257	***	ns	ns		
Dimethyl-sulfide	2.32^{b}	3.75^{a}	4.58^{a}	3.38^{a}	1.15^{b}	**	***	ns	-0.53	-0.24
Dimethyl-disulfide	35.9	56.3	28.0	56.5	53.8	*	*	ns		
Toluene	89.5^{b}	156^{b}	112	112	144	**	ns	ns		
Phenyl-Ethyl-alcohol	11.2^{a}	3.60^{b}	2.40^{b}	6.13^{b}	13.7^{a}	***	***	ns	0.64	0.37
Esterase activity										
Ethyl acetate	4.84^{a}	2.56^{b}	3.26	3.59	4.25	***	ns	ns	0.64	0.25
Ethyl pentanoate	3.99^{a}	0.96^{b}	1.71	2.23	3.50	***	*	*	0.81	0.34
Ethyl hexanoate	53.6^{a}	12.4^{b}	22.3	29.6	47.2	***	ns	ns	0.81	0.33
Ethyl heptanoate	1.39^{a}	0.28^{b}	0.42^{b}	0.72^{ab}	1.36^{a}	***	***	*	0.81	0.35
Ethyl octanoate	9.23^{a}	4.31^{b}	6.29	5.64	8.39	***	ns	ns	0.70	0.18
Ethyl decanoate	6.70^{a}	3.26^{b}	5.27	4.04	5.58	**	ns	ns	0.58	0.12
Packaging contaminants										
2-ethyl-hexene	12.1^{a}	6.47^{b}	11.2^{a}	7.08^{b}	9.53^{a}	***	**	ns	0.51	-0.26
BCA 1	40.4	72.4	39.1^{b}	101^{a}	28.5^{b}	*	***	ns	-0.64	0.54
BCA 2	5.38	8.18	5.11^{b}	11.4^{a}	3.80^{b}	*	***	ns	-0.63	0.67
BCA 3	3.40	4.56	3.46^{ab}	5.93^{a}	2.54^{b}	ns	***	ns	-0.59	0.62
BCA 4	5.7^{b}	12.5^{a}	3.01^{b}	20.8^{a}	3.57^{b}	**	***	ns	-0.60	0.67
BCA 5	47.8^{a}	35.2^{b}	28.4^{b}	67.5^{a}	28.4^{b}	**	***	**	0.07	0.61
BCA 6	40.6^{a}	13.2^{b}	14.3^{b}	30.1^{a}	36.5^{a}	***	***	*	0.79	0.49
BCA 7	15.6	31.1	4.71^{b}	61.7^{a}	3.73^{b}	ns	***	ns	-0.55	0.68
BCA 8	2.91	2.56	1.93^{b}	0.23	0.99^{b}	ns	***	ns	-0.12	0.50
BCA 9	3.20	5.62	1.27^{b}	9.52^{a}	2.43^{b}	ns	***	ns	-0.51	0.73
BCA 10	16.0^{a}	11.7^{b}	14.8	12.3	14.4	**	ns	ns	0.50	0.01
Unknow origin										
Acetal	2.57	2.85	2.72^{ab}	1.44^{b}	3.99^{a}	ns	**	ns		
Ethylbenzene	113	141	69.3^{b}	180^{a}	132^{a}	ns	***	ns		
p-xylene	111^{b}	155^{a}	83.9^{b}	178^{a}	137^{ab}	ns	**	ns	-0.50	0.39
m-xylene	39.0^{b}	67.8^{a}	35.4^{b}	70.9^{a}	53.9^{ab}	***	*	ns	-0.64	0.30
Cyclohexane	3.50	2.97	4.87^{a}	1.70^{b}	3.15^{ab}	ns	***	ns	0.15	-0.66
Butyl-cyclohexane	13.1	12.9	15.4^{a}	8.19^{b}	15.4^{a}	ns	***	ns	0.28	-0.61
3-Methyl-penthyl-cyclohexane	5.96	6.14	8.60^{a}	3.53^{b}	6.04^{ab}	ns	**	ns	0.19	-0.62

†Only VOCs with a significant effect were reported. Significant effect : * P<0.10; ** P<0.05 ; *** P<0.01. Estimated means within a row with different lower case letters are different (P<0.05).
††Only variable loadings ≥ ± 0.5 on PC1 or PC2 are reported.

The compounds originating from the oxidative decomposition of lipids were most abundant in the Parma hams; oxidation compounds proved to be increased by PLA packaging. Dry-cured ham nature accounted for difference in compounds from carbohydrate fermentation. VOCs produced from amino acid catabolism were detected mostly in Teruel hams and were increased by PLA packaging. According to these results, the "breathable" PLA plays an effective role in enhancing both oxidation and maturation mechanisms. Ethyl esters were a feature of P hams, formed from free fatty acids with the high amount of ethanol found in these hams. Branched chain alkanes (BCAs) could not be identified with the available libraries and were named with subsequent numbers. These compounds increased significantly in PET packaging, as a possible consequence of migration from packaging material in direct contact with the ham slices, favoured by fat content (range 10-18% on wet slice) and high surface/volume ratio. Also ethylbenzene, m- and p-xylene increased in PET packaging. Principal Components Analysis (PCA) was carried out including VOCs listed in Table 1, and the scores of fresh (F) and packaged samples were plotted onto the PC1-PC2 plane (Fig. 1). P-PET and T-PET packaged hams differed from the fresh ones (P-F and T-F) along PC2, mainly discriminated by the BCAs content (Table 1). PLA packaged P (P-PLA) hams differed remarkably from the fresh P hams along PC1 and PC2 (increase of oxidation compounds, see Table 1). F and PLA packaged T samples are grouped closely, showing the stability of T hams in PLA.

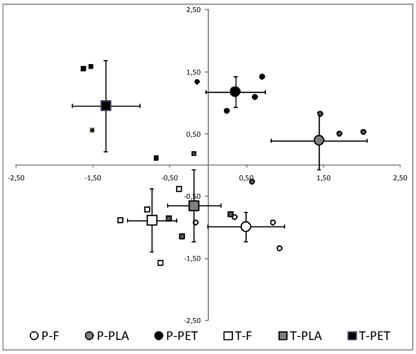

Fig. 1. Scoreplot of fresh and packaged sliced dry-cured hams. Large symbols represent the mean scores of each ham group, small symbols represent single samples, thin bars represent standard deviations of PC1-PC2 scores within groups.

IV – Conclusions

VOCs of Parma and Teruel dry-cured hams showed remarkable differences as a consequence of different processing way and formulation. Packaging in PLA material stressed differences between ham types, mainly due to the sharp increase of compounds generated by lipid oxidation in Parma ham. The effect of PET packaging was the rise of BCAs in both ham types, as a possible consequence of migration from packaging material in direct contact with the product. The improvement of safety and quality of ready-to-eat packaged dry-cured meat products will require further research on packaging materials.

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Meat quality traits of heavy pigs for the production of traditional Tuscan salami

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Abstract. The objective of the study was to evaluate the effect of the replacement of corn meal (CoM) by chestnut meal (ChM) in two diets of heavy pigs on the quality of meat and traditional Italian Salami obtained. Sixteen Large White pigs were divided in two groups and reared indoors in Garfagnana Valley (Tuscany, Italy). All animals were slaughtered after 464 days of trial at about 228 kg live weight. The quality of the *Longissimus lumborum* muscle, subcutaneous backfat and salami sausages was evaluated by chemical analysis and fatty acids profile. Data obtained show that ChM group increased the incorporation of unsaturated and monounsaturated fatty acids in muscle and backfat ($P<0.05$), moreover the diet with ChM significantly reduce ($P<0.05$) saturated fatty acids in backfat. Particularly ChM diet produced an increase ($P<0.05$) in the content of C18:1n−9 and significantly ($P<0.05$) reduce the content of C16:0, C18:0 and C20:0 in backfat. Inclusion of chestnut in the diet seems to have no significant effect on the fatty acid profile of salami. It is concluded that feeding a diet with ChM instead of CoM altered the fatty acid composition of *Longissimus lumborum* muscle and backfat without simultaneously affecting characteristics of salami sausages quality.

Keywords. Heavy pigs – Chestnut – Meat quality – Salami.

Qualité de la viande de porc lourd pour la production de charcuterie traditionnelle de la Toscane

Résumé. *L'objectif de cette étude était d'évaluer l'effet du remplacement de la farine de maïs (CoM) par de la farine de châtaignes (ChM) dans les régimes de porcs lourds, sur la qualité de la viande et du salami italien traditionnel obtenu. Seize porcs Large White ont été divisés en deux groupes et élevés à l'intérieur de la vallée de la Garfagnana (Toscane, Italie). Tous les animaux ont été abattus après 464 jours avec un poids d'environ 228 kg de poids vif. La qualité du muscle* Longissimus lumborum*, du lard et des salami traditionnels a été évaluée par analyse chimique et par profil en acides gras. Les résultats obtenus montrent pour le groupe ChM un niveau plus élevé en acides gras insaturés et monoinsaturés chez le muscle et le lard (P<0,05), en plus, le régime à base de ChM a conduit à une diminution significative (P<0,05) des acides gras saturés du lard. Particulièrement ChM a conduit à une augmentation (P<0,05) du contenu de C18:1n-9 tout en baissant (P<0,05) le niveau de C16:0, C18:0 et C20:0 du lard. L'inclusion de la châtaigne dans le régime ne semble pas avoir d'effet sur le profil en acides gras du salami. Nous avons conclu qu'un régime ChM au lieu de CoM modifie la composition en acides gras du muscle et du lard, sans pour autant avoir des interférences sur les caractéristiques de la qualité des salami.*

Mots-clés. *Porc lourd – Châtaignes – Qualité de la viande – Salami.*

I – Introduction

In the territory of Garfagnana (Lucca, Tuscany) pig rearing is carried out for processing meat into high quality traditional sausages (Register of traditional Tuscan products), the technique of production involves the use of animals slaughtered at high live weights (over 200 kg) and fed with by-products of the district derived from the processing of chestnut fruit (*Castanea sativa*) and spelt grains (*Triticum dicoccum*) for human food use. The high availability of these products allows local farmers to reduce feed costs and to typify the processed products.

These feed resources, especially the chestnut meal, confer specific dietary characteristics to meat by changing the lipid fraction increasing the proportion of unsaturated fatty acids (Coutron-Gambotti *et al*., 1998; Pugliese *et al*., 2005). Pigs used in this rearing system belong to cosmopolitan breeds indeed carcass traits are highly suitable for the processing into sausages.

The objective of this research was to evaluate the effect of substitution of corn meal with chestnut meal on chemical composition and fatty acids profile of muscle, backfat and a typical Tuscan salami derived from heavy pigs.

II – Materials and methods

The trial were carried out in a farm located in the Garfagnana district on 16 Large White reared intensively and slaughtered at around 228 kg live weight. In the growing period pigs were fed with a mixed diet (spelt meal, spelt bran, corn meal, soybean meal), while during the finishing period (95 days), heavy pigs were divided in two groups (Table 1) and fed with integration of corn meal (CoM) or chestnut meal (ChM).

Table 1. Composition of the experimental diets (%)

	Soybean meal	Corn meal	Chestnut meal	Spelt meal	Spelt bran
CoM†	6	44	0	38	12
ChM††	6	34	10	38	12

†CoM corn meal; ††ChM chestnut meal.

At the slaughtering, samples of muscle and backfat from each group were taken and stored for the chemical analysis. Traditional *Salame nostrano* were prepared by a local sausage make based from two diets. The lean meat (80%) were obtained from the sizing of ham, shoulder, loin, fillet, cup and fat part (20%) from backfat, cheek, shoulder. Minced into small pieces, the meat was mixed with salt (2.5%), black pepper (0.5%), infusion of garlic, nutmeg and white wine; no additives, sugar or starter were used. The meat was cut into a small pieces and were mixed for obtained a traditional grinding (7 mm). The dry sausages was made by means of a mechanical pressured sausages making machine and packed into natural gut casings (soft pork intestines) and hand-tied. Traditional salami after drying period (7 days) and ripening period (53 days) were prepared; the traditional room temperature ripening ranged from 7°C to 18°C, while the relative humidity ranged from 75% to 80%. At the end of experiment all the salami were weighed an average of about 550 g.

A representative samples of salami were vacuum packaged and stored at -20°C until chemical analysis. On the samples proximate chemical composition were carried out according to the AOAC method (1990), the extraction of total lipid was determined as described by Folch (1957), fatty acids methyl esters were prepared according to the method described by Christie (1982). FAMEs were analyzed by gas chromatography using a Thermo Quest (Milan, Italy) GC apparatus equipped with a 100-m high polar fused silica capillary column and a flame ionization detector (i.d. 0.22 mm, 0.25 μm film thickness; Chrompack CP-Sil 88 Varian, Middelburg, The Netherlands). Separated FAMEs were identified by comparison with the retention times of pure standards and reported as percentages of total fatty acids. Data were analyzed by one-way analysis of variance with JMP7 software (SAS Institute). Statistical significance was established at the level of $P<0.05$ and comparisons between means were conducted using the Tukey's HSD test.

III – Results and discussion

Results of the proximate chemical composition of diets are presented in Table 2 that were formulated as isoenergetic and isoprotein. CoM diet has a higher content of PUFA, particularly C18:2n-6 and C18:3n-3 while ChM showed a higher content in C18:0 and C18:1n-9 in agreement with other research on Corsican pig (Coutron-Gambotti *et al*., 1998) where the chestnut has increased the content of oleic acid in the diet.

Table 2. Chemical analysis (% DM) and major fatty acid composition of diets (% total FA)

	CoM	ChM
Moisture	9.27	9.06
Crude protein	14.04	13.88
Crude fat	2.72	2.66
Crude fiber	4.89	5.00
Ash	2.39	2.57
DE†	15.10	14.90
C14:0	0.04	0.15
C16:0	13.07	12.63
C16:1n-7	0.17	0.40
C18:0	1.28	1.68
C18:1 n-9	24.70	26.64
C18:2 n-6	54.95	52.93
C18:3 n-3	3.51	2.57
C20:0	0.21	0.35
C20:1 n-9	0.70	0.69
C22:0	0.18	0.20
C24:0	0.06	0.21
SFA	14.95	15.37
UFA	85.05	84.56
PUFA	58.46	55.60

† Digestible Energy expressed as MJ/kg DM.

No statistical differences were observed for the proximate analysis in muscle, backfat and salami among experimental diets (Table 3). The results for S*alame nostrano* were comparable with the data reported in the literature of Italian salami (Moretti *et al*., 2004).

The fatty acid composition of muscle in pigs fed with chestnut meal showed higher values of MUFA and PUFA (C18:3n-3) according to other research carried out on Cinta Senese and Iberian pig breeds (Pugliese *et al*., 2005; Andrés *et al*., 2001; Cava *et al*., 1999). The fatty acid composition of backfat showed a high values in SFA as a result of higher content in C14:0, C16:0, C18:0, C20:0 in CoM diet, while ChM significantly modified MUFA, particularly oleic acid and the PUFA/SFA ratio. The chestnut meal led to a higher content of C18:3n-3, C20:2n-6 and C20:4n-6 fatty acids, as reported previously by other authors (Diaz *et al*., 1996). In traditional salami obtained from pig fed with CoM diet the results showed a high level of cis-vaccenic (C18:1n-7) and linoleic (C18:2n-6) acid probably due to the high content of corn meal in diet.

IV – Conclusions

The integration of chestnut flour in the diet of heavy pig can affect the fatty acid profile of fresh products (backfat and muscle) by increasing the content of unsaturated fatty acids and particularly MUFA. In the processed product (salami) results show that the addition of small amounts of chestnut flour involves minor effects on the lipid fraction.

This research shows that the use of chestnut flour and other by-products of the area can improve the quality aspect and identify local meat products, moreover the use of these feed can help to reduce costs and represent an economic advantage for the farmer.

Table 3. Proximate analysis (%) and fatty acid composition of muscle, backfat, salami (% total FA)

	Muscle			Backfat			Salami		
	CoM	ChM	SE	CoM	ChM	SE	CoM	ChM	SE
Moisture	70.25	70.62	0.55	96.25	95.86	0.32	24.96	25.28	1.20
Crude Protein	76.40	76.28	1.88	-	-	-	38.98	35.28	1.43
Total Lipids	2.79	2.78	0.55	40.56	47.07	0.42	21.05	19.70	0.36
Ash	3.54	3.69	0.10	-	-	-	5.52	5.21	0.17
C10:0	0.18[a]	0.16[b]	0.01	0.08	0.09	0.01	0.08	0.07	0.01
C14:0	2.09	2.13	0.08	1.89	1.98	0.11	1.37	1.27	0.06
C16:0	21.78	21.88	0.40	22.16[a]	20.62[b]	0.36	22.59	23.08	0.27
C16:1 n-7	3.68	4.43	0.39	1.89	1.84	0.36	2.52	2.61	0.08
C18:0	10.54	9.31	0.56	12.25[a]	10.53[b]	0.51	11.67	11.81	0.30
C18:1 n-9	43.93	45.36	0.76	40.32[a]	42.62[b]	0.94	43.44	43.52	0.40
C18:1 n-7	4.66	5.25	0.30	2.49	2.51	0.30	3.13[a]	3.22[b]	0.10
C18:2 n-6	8.21	7.12	0.75	13.65	14.46	0.92	10.56[a]	10.02[b]	0.12
C18:3 n-3	0.26[a]	0.28[b]	0.22	0.64[a]	0.74[b]	0.02	0.51	0.47	0.01
C20:0	0.20	0.19	0.03	0.25[a]	0.20[b]	0.01	0.21	0.22	0.01
C20:1 n-9	0.75	0.92	0.16	1.20	1.15	0.15	1.02	1.02	0.01
C20:2 n-6	0.34[a]	0.33[b]	0.03	0.79[a]	0.84[b]	0.02	0.56	0.52	0.01
C20:4 n-6	1.10	0.60	0.15	0.17[a]	0.23[b]	0.20	0.21	0.21	0.01
C22:4 n-3	0.15	0.14	0.02	0.09	0.10	0.02	0.07	0.08	0.06
C22:5 n-3	0.10	0.06	0.03	0.02	0.05	0.03	0.05	0.04	0.01
SFA	35.79	33.67	1.38	36.85[a]	34.40[b]	0.59	36.50	37.04	0.49
UFA	63.67[a]	65.48[b]	1.34	63.03[a]	65.46[b]	0.58	63.38	62.85	0.48
MUFA	53.52[a]	56.96[b]	1.65	46.65[a]	49.38[b]	0.63	51.24	51.35	0.55
PUFA	10.15	8.52	1.38	15.70	16.81	0.72	12.22	11.60	0.16
PUFA/SFA	0.28[a]	0.25[b]	0.02	0.43[a]	0.49[b]	0.05	0.33	0.31	0.05
n-6/n-3	19.14	16.94	3.76	18.51	16.45	0.58	16.67	17.19	0.47

a, b within criterion means different ($P<0.05$).

Acknowledgements

The project was supported by grant of Fondazione Cassa di Risparmio di Lucca. The authors thank to technical staff of Antica Norcineria, Ghivizzano (Lucca).

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Colour stability during prolonged storage of dry fermented sausages from Iberian pork

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Abstract. 30 units of chorizo and 30 units of salchichón "sarta" were packaged in different gas atmosphere conditions (i) vaccuum packaging (Batch 1) (ii) 70% N_2+ 30% CO_2 (Batch 2) (iii) 100 N_2 (Batch 3) (iv) 70% Argón + 30% CO_2 (Batch 4), and were stored under refrigeration (4±1ºC) during nine months. The evolution of several parameters related to colour surface (L*, a*, b*, Croma and Hue angle) were studied all throughout storage period. There were no differences for luminosity (L*) among packaging treatments for chorizo and salchichón samples (P>0.05). Red colour intensity (a*) significantly decreased for chorizo "sarta" in every batch, from initial values of 16.89±0.53 to final values ranging 14.17-15.22±0.55-0.49. With respect of differences among packaging systems of chorizo, batch 4 (70% argón+30% CO_2) showed the highest a* and C-values after 180 days and batch 2 (70% N2 + 30% CO2) the lowest, indicating more intense oxidation reactions affecting the pigment of samples packed in this gas composition. However, these differences were not evident after 270 days of storage. With respect of salchichón "sarta", a decrease in redness was evidenced alter 180 days of storage as well as a reestablishment of red colour at the end of the storage period batch 2 (70% N_2+30% CO_2) being the batch with the highest red colour intensity (P<0.001). The results obtained in this study suggest that as far as external appearance, specially colour, is concerned, shelf life of chorizo and salchichón "sarta" could be even longer. With respect of the type of gas atmosphere, this factor was not relevant for chorizo, whereas for salchichón, the gas atmosphere consisting in 70% N_2+30% CO_2 was the most convenient.

Keywords. Chorizo "sarta" – Salchichón "sarta" – Modified atmosphere packaging – Colour.

La stabilité de la couleur durant le stockage prolongé des charcuteries crues affinées de porc Ibérique

Résumé: *30 unités de "chorizo" et 30 de saucisson du type "sarta" ont été emballées sous différentes atmosphères : (i) emballage sous vide (Lot 1), (ii) 70% N_2+ 30% CO_2 (Lot 2), (iii) 100% N_2 (Lot 3), (iv) 70% Argon + 30% CO_2 (lot 4), et elles ont été ainsi stockées sous réfrigération (4±1ºC) pendant 9 mois. L'évolution des paramètres en rapport à la couleur de la superficie a été également étudiée (L*, a*, b*, Chrome et Hue) pendant la période de stockage. Il n'y a pas eu de différences de luminosité (L*) dues aux différentes compositions en gaz dans l'emballage utilisé pour les échantillons de "chorizo" et de saucisson du type "sarta" (P>0.05). Dans le cas du "chorizo sarta" l'intensité de la couleur rouge (a*) s'est significativement réduite dans tous les lots, depuis les valeurs initiales de 16,89±0,53 jusqu'aux valeurs finales de 14,17-15,22±0,55-0,49. En ce qui concerne les différences dues à la composition en gaz de l'emballage utilisé pour le "chorizo", le Lot 4 (70% argon+30% CO_2) a présenté les valeurs les plus élevées pour a* et C, 180 jours après, et le Lot 2 (70% N_2 + 30% CO_2) les moins élevées en montrant une oxydation plus intense de la pigmentation dans les échantillons emballés dans ce gaz. Néanmoins, ces différences n'étaient pas perçues après 270 jours de stockage. Concernant le saucisson "sarta", on observe une réduction de l'intensité de la couleur rouge après 180 jours d'emballage et sa récupération ultérieure vers la fin de la période d'emballage, le Lot 2 (70% N_2+30% CO_2) étant celui qui a présenté une plus grande intensité de couleur rouge dans le produit (P<0,001). Ces résultats nous suggèrent qu'en ce qui concerne l'aspect déterminé par la couleur, la durée de vie utile du "chorizo" et du saucisson du type "sarta" pourrait se prolonger plus longtemps. Et concernant le type d'emballage, ce facteur n'a pas exercé d'effet significatif dans le cas du "chorizo", et dans le cas du saucisson, le mélange 70% N_2+30% CO_2 a été le plus intéressant.*

Mots clés: *Chorizo "sarta" – Saucisson "sarta" – Emballage en atmosphères modifiées – Couleur.*

I – Introduction

Salchichón and chorizo are the most popular fermented raw cured meat, existing a great variety of them depending on the area of production (Edwards *et al*., 1999). The kind of variety "sarta" shows a diameter bigger than 22 millimetres and the sausage has got a horseshoe shape.

Currently, it is a more and more frequent practice in the packing of these raw fermented products vacuum or in different gas atmospheres (packed in modified or protector atmospheres, EAM or EAP) with the aim of adapting the sector to the new demands and consumption tendencies. The EAM consists in the replacement of the air that surrounds the food, by a gas or more frequently by an optimum mixture of gases that permits the lengthening of it shelf-life.

The shelf-life of fermented raw cured meat products is determined by their appearance and especially by the colour they show. During the curing process of these products the myoglobin transforms into nitrosylmyoglobin and nitrosylhemocrome, which are more stable pigments than myoglobin. Nevertheless the colour of fermented raw meat products can be altered during the cold storage (Ruiz Pérez-Cacho *et al.*, 2005). The effect of the packed on these characteristics has been studied in other works (Fernández-Fernández *et al.*, 2002). For that reason the aim of this study was to determine the most convenient packing conditions for the optimum preservation of the colour of the chorizo and salchichon samples of the "sarta" kind during a cold storage of 9 months.

II – Material and methods

For the development of this study 60 (30 and 30) units of chorizo and salchichón "sarta" that weighted between 0.261 ± 0.0089 and 0.224 ± 0.0111 kg respectively were used. These products were manufactured by Montesano SA company according to the standard formulation. Both kinds of sausage present a standardized amount of fat and lean (30% and 70% respectively). The period of ripening was concluded when the decrease of both products reached a 35-37%.

Each unit of product was packed in a bell packaging of the brand Tecnotrip, mod. EV-13-CB, Nº 932334 for the vacuum packing lots, an ULMA brand Flow-Pack machine mod. PV 350 LSHIX EMB and Nº 1219098 for the rest of the lots and in different conditions: (i) vacuum (Lot 1), (ii) 70% N_2+ 30% CO_2 (Lot 2), (iii) 100 N_2 (Lot 3), (iv) 70% Argon + 30% CO_2 (Lot 4). The vacuum plastic material consisted in PA/PE 30/120 of 150 microns of thickness with an O_2 of 25-30 cm^3/m^2/bar/24h at 23ºC permeability range and a water vapour transmission of 1,7 g/m^2/24 h at 23ºC and 85% of relative humidity. The samples were stored in refrigeration for 9 months, taking samples in the beginning, after 6 months and in the end of the storing period.

The colour of the surface cut in the chorizo and salchichón "sarta" was immediately measured after the opening of the pack by three times according to the American Meat Science Association (AMSA, 1991) recommendations. The following colour coordinates were obtained: luminosity (*L**), red colour intensity (*a**, red ± green) and yellow colour intensity (*b**, yellow ± blue). *a** and *b** were used to calculate the hue angle (hue = arctan [*b**/*a**]) and the colour saturation (chroma= $[a^*2 + b^*2]^{0.5}$) parameters. The data obtained were analysed by the SPSS (SPSS 13.0) software package.

III – Results and discussion

The Tables 1 and 2 present the evolution of the colour parameters instrumentally measured (*L*, a*, b**, colour saturation or C and angle of Hue or h). The values of initial luminosity in the case of chorizo and salchichón "sarta" were of 32,84±1,01 and 32,37±0,94 respectively. During the first 180 days of storage it is possible to observe in a general way that the luminosity values were reduced in comparison to the initial ones, in some cases in a statistically significant way

(P<0.05) (Lot 2:70% N_2+30% CO_2 and Lot 3: 100 N_2 for the salchichón and lot 1: vacuum, Lot 2: 70% N_2+30% CO_2 and Lot 3: 100 N_2 for the chorizo). However, after 270 days of storage it is showed that the values of luminosity increase again in the mentioned lots, even if they do not reach the initial values of luminosity registered. In a similar way Rubio *et al.*, (2007) observed that the values of luminosity in samples of salchichón stored for 210 days were even higher in comparison with the initial product (P<0.05).

According to the differences among packed lots, these weren't significant (P>0.05) excepting the chorizo samples after 180 days of storage presenting the lot 3 (100% N_2) the maximum values of luminosity and the lot 2 (70% N_2 30% CO_2) the minimum. According to the evolution of the red colour in the different studied products, the intensity of red colour (*a**) was significantly reduced in the case of chorizo "sarta" in every lot, from initial values of 16.89±0.53 to final ones of 14.17-15.22±0.55-0.49. The loss of intensity in red colour is mainly owed to the oxidation of nitrosylmyoglobin an the formation of metmyoglobin, due to the presence of residual oxygen quantities (Andersen and Skibsted, 1992).

Table 1. Instrumental colour evolution (L*, a*, b*, C, h) (mean± standard error of the mean) on chorizo "sarta" stored under modified atmosphere packaging at 4ºC during 9 months

Time (days)	Packaging*	L* ± SE		a* ± SE		b* ± SE		C ± SE		H ± SE	
0		32.84$^{1/1/1/-}$	1.01	16.89$^{1/1/1/1}$	0.53	10.22$^{1/1/-/-}$	0.55	19.77$^{1/1/1/-}$	0.71	30.83$^{-/-/-/1}$	0.9
180	1	29.17ab^{2}	0.45	14.46ab^{2}	0.39	8.86ab^{2}	0.29	16.99ab^{2}	0.45	31.5	0.6
	2	28.57b^{2}	0.38	13.38b^{3}	0.34	8.24b^{2}	0.3	15.73b^{3}	0.43	31.48	0.52
	3	30.66a^{12}	0.53	14.05ab^{2}	0.35	9.32ab	0.37	16.90ab^{2}	0.46	33.24	0.78
	4	30.54a	0.51	15.04a^{12}	0.54	9.99a	0.47	18.08a	0.7	33.311	0.55
	$P_{envasado}$	***		*		*		*		NS	
270	1	31.2412	0.63	15.222	0.49	9.1812	0.4	17.792	0.61	30.94b	0.67
	2	31.791	0.61	14.802	0.33	9.611	0.28	17.662	0.41	32.90a	0.5
	3	30.012	0.7	14.172	0.55	9.01	0.53	16.822	0.73	31.96ab	0.67
	4	30.13	0.48	14.332	0.33	9.44	0.3	17.18	0.43	33.17a^{1}	0.39
	$P_{envasado}$	NS		NS		NS		NS		*	

Lot 1 = (vacuum); Lot 2= (70% N_2+ 30% CO_2); Lot 3=100 N_2; Lot 4= 70% Argon + 30% CO2. Significance levels: ns=>0.05:*=*p*<0.05: **= *p*<0.01: ***= *p*<0.001. a,b,c: different letters in the same column within the same storage time, mean significant differences between lots (P<0.05).
Test of Tukey 1,2,3: different superscripts in the same column within the same lots, mean significant differences between storage time (P< 0.05). Test of Tukey

Regarding the differences related to the gas composition of the pack, the lot 4 (70% argón+30% CO_2) showed the maximum values of *a** and C after 180 days and the lot 2 (70% N_2 + 30% CO_2) the minimum, pointing a more intense composition of the pigment in the samples packed in this gas. Nevertheless, these differences weren't perceptible after 270 days of storage.

Regarding the salchichón "sarta", a reduction in the intensity of red colour is observed after 180 days of packing and the following recovering of it in the end of the storage, being the Lot 4 (70% argon+30% CO_2) the one that showed a mayor intensity of red colour in the product after 270 days of storage(P<0.001). The loss of intensity in red colour is owed to the formation of metmyoglobin (of brown colour) from the nitrosylmyoglobin (MbFe(II)NO) (Lindahl *et al*, 2001). The reduction of intensity in red colour and the following recovering of it have been observed in previous studies in dry-cured ham (Andrés *et al.*, 2005) as well as in cured-fermented product (Rubio *et al*., 2008). Andersen *et al.* (1998) also observed that the mentioned recovering of colour was more important if an adecuate exclusion of oxygen in the pack was performed. On the other hand, it can be supposed that the loss of humidity in the product during it storage can be related.

Table 2. Instrumental colour evolution (L*, a*, b*, C, h) (mean± standard error of the mean) on salchichón "sarta" stored under modified atmosphere packaging at 4 ºC during 9 months

Time (days)	Packaging*	L* ± SE		a* ± SE		b* ± SE		C ± SE		H ± SE	
0		$32.37^{-/1/1/-}$	0.94	$11.35^{1/1/1/1}$	0.22	$4.01^{1/1/1/1}$	0.11	$12.04^{1/1/1/2}$	0.23	$19.45^{2/2/2/2}$	0.34
180	1	30.86	0.5	8.783	0.17	3.162	0.07	9.333	0.18	19.802	0.25
	2	29.792	0.39	8.333	0.17	2.982	0.08	8.863	0.17	19.792	0.54
	3	29.982	0.46	8.543	0.13	3.033	0.07	9.063	0.15	19.502	0.25
	4	30.43	0.64	8.312	0.16	3.003	0.08	8.843	0.17	19.852	0.38
	$P_{envasado}$	NS		NS		NS		NS		NS	
270	1	30.74	0.54	$9.59ab^{2}$	0.15	$3.80a^{1}$	0.09	$10.32ab^{2}$	0.17	$21.57b^{1}$	0.41
	2	31.0412	0.42	$9.82a^{2}$	0.15	$3.82a^{1}$	0.07	$10.54a^{2}$	0.16	$21.26b^{1}$	0.27
	3	30.9512	0.51	$9.21b^{2}$	0.15	$3.64a^{2}$	0.08	$9.91b^{2}$	0.16	$21.59b^{1}$	0.39
	4	31.77	0.5	$8.71c^{2}$	0.12	$3.36b^{2}$	0.08	$9.34c^{2}$	0.12	21.081	0.45
	$P_{envasado}$	NS		***		***		***		NS	

Lot 1 = (al vacío); Lot 2 = (70% N2+ 30% CO2); Lot 3 = 100 N2; Lot 4 = 70% Argon + 30% CO2. Significance levels: ns =>0.05:*=p<0.05: **= p<0.01: ***= p<0.001. a,b,c: different letters in the same column within the same storage time, mean significant differences between lots (P<0.05).
Test of Tukey. 1,2,3: different superscripts in the same column within the same lots, mean significant differences between storage time (P< 0.05). Test of Tukey

Chroma or saturation of colour follows similar evolution to the one explained for *a** in salchichón and chorizo, being significantly reduced during the first 180 days of storage and increasing afterwards during the end of the period of storage, although if it doesn't reach the initial values of 12.04±0.23. The mayor value of Chroma on the surface of salchichón from lot 4 (70% argón+30% CO_2) has to be remarked comparing to the rest of the lots. This parameter represents the brightness of colour on the surface of the product (Sarasibar *et al.*, 1989). *b** is a parameter normally related to the lipid oxidation. In the case of chorizo "sarta" results hardly affected by the time of storage and only after 180 days, differences owed to the mixture of used gases are observed, showing the Lot 4 (70% argón+30% CO_2) the maximum value. Other authors have observed that the variation of *b** in this product cab be related to the presence of carotenoids (β-caroteno and criptoxantina) included in the páprika pepper, typical spice used in chorizo (Gimeno *et al.*, 2000). The nitrificant salts reduce the intensity and stability of the paprika developing a yellow discoloration of the red paprika of low pH (Sarasibar *et al.*, 1989). In the case of salchichón "sarta" the maximum values of *b** have to be remarked in lot 4 at the end of the period of storage, because they can point a mayor lipid oxidation.

IV – Conclusions

The results obtained in this study suggest that regarding the appearance determined by the colour, the-shelf life of the chorizo and salchichón "sarta" could be extended for a longer time. Regarding the packing time, this factor didn't exert a significant effect in the case of chorizo, and in the case of salchichón, the mixture 70% argon+30% CO_2 was more convenient.

Acknowledgements

The authors of the article are grateful to Junta de Extremadura for the concession of the project and to Montesano SA because they have make possible the execution of this study, and finally to Fundecyt for the management conducted.

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A further look on genetic basis of carcass fat deposition in pigs of 'Casertana' ancient autochthonous genetic type

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Abstract. 'Casertana' pig ancient autochthonous genetic type (AAGT) has been considered in the years within wide programme of safeguard and rescue promoted by ConSDABI. One of the most relevant aims of this program is to search for gene variation related to quantity and technological, nutritive and organoleptic quality of fat in order to satisfy consumer's request. In the present work, 100 AAGT Casertana pigs were genotyped for 8 SNP at *loci* involved in the regulation of adipose tissue deposition [DECR1, FASN, MC4R, SCD and H-FABP haplotype (H-FABP1, H-FABP2, H-FABP3)] in order to carry out an association study. with some fatness characteristics of carcass. Statistical analysis was performed using the GLM procedure of the SAS package. The results showed association between HFABP haplotype and the majority of considered phenotypic traits. In detail, the effect of this haplotype was significant on the total weight of separable fat in carcass (P=0.002), total weight of adipose cuts (P=0.006) and back fat thickness measured at level of 1st thoracic vertebra (Th1) (P=0.056), 15th thoracic (Th15) (P= 0.020) and between the 6th lumbar vertebra and sacrum (L6 – S) (P=0.007). FASN showed an effect on belly and jowl fat (P≤0.05). For DECR1 CC genotype influenced the weight of belly cut. Further investigations are ongoing for an operative utilisation of the H-FABP haplotype, FASN and DECR1 genes as molecular markers (candidates) in proper molecular assisted selection (MAS) plans.

Keywords. Casertana 'AAGT' – Fat traits – SNPs – Haplotype.

Une perspective sur les bases génétiques du dépôt de graisse dans la carcasse des porcs du type génétique autochtone ancien 'Casertana'

Résumé. *Le porc 'Casertana', type génétique autochtone ancien (TGAA) du "bioterritorio" de la Campanie, est depuis des années l'objet d'un programme de protection et de valorisation mis en œuvre par le ConSDABI SUB NFP.I - FAO. Parmi les objectifs de ce programme il y a celui d'identifier des variantes des gènes associés à la quantité et à la qualité technologique, nutritionnelle et organoleptique des matières grasses pour répondre aux besoins du consommateur. Dans cet article, nous avons étudié 8 SNP dans les loci candidats au déterminisme quantitatif du tissu adipeux [DECR1, FASN, MC4R, SCD et haplotype H-FABP (H-FABP1, H-FABP2, H-FABP3)]. 100 porcs TGAA 'Casertana' pour lesquels certains relevés étaient disponibles concernant des sections, ont été génotypés afin d'effectuer une étude d'association avec quelques caractéristiques de l'adiposité. L'analyse statistique a été réalisée en utilisant la procédure GML du SAS. Les résultats suggèrent une association entre l'haplotype H-FABP et la plupart des caractères en question. En particulier, l'effet de l'haplotype H-FABP est significatif sur le poids total des morceaux de gras (P=0,006), sur les gras séparables (P =0,002) et sur l'épaisseur de gras mesurée à la 1ère vertèbre thoracique (Th1) (P=0,056), à la 15ème vertèbre thoracique (Th15) (P= 0,020) et entre la 6ème vertèbre lombaire et le sacrum (L6 – S) (P=0,007). En outre, FASN a des associations avec le lard de la poitrine et le lard de la bajoue (P ≤ 0,05). D'autres recherches sont en cours pour une utilisation opérationnelle des haplotypes H-FABP et des gènes FASN et DECR1 comme marqueurs prometteurs ('candidats') dans les plans de sélection assistée par marqueurs moléculaires (MAS).*

Mots-clés. *TGAA 'Casertana' – Caractéristique de l'adiposité – SNP – Haplotype.*

I – Introduction

'Casertana' (CT) ancient autochthonous genetic type (AAGT) is a black pig of ancient origins. It is object of considerable interest for its many peculiarities, like: good aptitude to grazing with ability to utilize poor feed; appreciable organoleptic and healthy quality of meat which is particularly suitable to obtain valuable local products (Matassino *et al*., 1968; Colatruglio *et al*., 1994; Girolami *et al.,* 1996; Matassino *et al*., 2006; Barone *et al*., 2008). In the last years, the awareness of the nutritional issue has increased: healthy quality is the element that more concerns the consumer. In this context, the study of quanti-qualitative characteristics of the adipose tissue as well as of the factors influencing them in farm animals, especially of pig, has become increasingly important. Furthermore ongoing research at ConSDABI SUB NFP.I - FAO on pig AAGTs is corroborating the hypothesis to consider pig as a valid model to study human obesity. These pigs, characterized by a thicker back fat than cosmopolite breeds, can constitute an interesting resource to contribute to the knowledge of genetic factors involved in obesity; moreover, AAGTs can constitute a genetic reserve suitable to rescue organoleptic properties penalized in cosmopolite breeds. Different approaches are used to identify molecular markers linked to traits associated to adipose tissue deposition in pig: (i) candidate gene; (ii) QTL identification; (iii) combination of (i) and (ii); (iv) transcriptome analysis (Davoli *et al*., 2009). Various genes involved in quali-quantitative characteristics of muscle and adipose tissue have been identified. The aim of the present contribute was to evaluate, in CT pig, possible associations between 8 SNPs at *loci* involved in regulation of adipose tissue deposition [DECR1, FASN, MC4R, SCD and H-FABP haplotype (H-FABP1, H-FABP2, H-FABP3)] and some fatness characteristics of carcass; these *loci* are known in literature for their significant association with some fatness traits (Gerbens *et al*., 1997; Kim *et al*., 2000; Wimmers *et al.,* 2002; Munoz *et al*., 2003; Amills *et al*., 2005; Matassino *et al*., 2007 and 2009).

II – Materials and methods

DNA was extracted from blood and muscle samples of 100 subjects of CT AAGT, reared at experimental Farm of ConSDABI SUB NFP.I - FAO. Genotyping for SNPs considered at DECR1, FASN, H-FABP, MC4R and SCD *loci* was carried out by PCR-RFLP method according to literature protocols (Table 1).

Table 1. SNPs investigated in 'candidate' *loci* object of study

SNP	*LOCUS*				BIBLIOGRAPHIC REFERENCE
	ACRONYM	DENOMINATION	FUNCTION	CHROMOSOME	
C(163)G	DECR1	2,4 - Dienoyl reductase 1 mithocondrial	Encodes for an enzyme involved in unsaturated fatty acid beta-oxidation	4	Davoli *et al*., 2002
T265C	FASN	Fatty acid synthase	Enzyme key in the conversion of acetyl-CoA and malonyl-coA into long-chain satured fatty acids	12	Munoz *et al*., 2003
C(1811)G H-FABP1 (D/d) T(1324)C H-FABP2 (H/h) T(1970)C H-FABP3 (A/a)	H-FABP	Heart fatty acid - binding protein	Trafficking of fatty acids at level of cardiac and striate muscle as well as lactating mammary gland	6	Gerbens *et al*., 1997
G(298)A	MC4R	Melanocortin 4 receptor	Control of uptake capacity and growth rate	1	Kim *et al*., 2000
T(233)C SCD1 C(641)T SCD3	SCD	Stearoyl CoA Desaturase	Encodes for an enzyme key in the monounsaturated fatty acid biosynthesis	14	Ren *et al.*, 2004

For each SNP the allelic and genotypic frequencies were estimated. Concerning H-FABP, 12 out of 27 possible haplotypes were individuated, only 6 of which (HHddAa, HHddAA, HHDdAa, HHDdAA, HHddaa, HhDdAa) were used for statistical elaboration because of the insufficient numerousness of the others; for each fatness indicator 15 possible pair-wise comparisons were carried out. In the present contribute the following parameters at carcass dissection were considered as adiposity indicators: (i) total weight of adipose cuts (with and without 'jowl'); (ii) total weight of separable fat; (iii) thickness (mm) of back fat measured at level of: 1^{st} thoracic vertebra (Th1), 15^{th} thoracic vertebra (Th15) and between the 6^{th} lumbar vertebra and sacrum (L6 - S).

Statistical elaboration, concerning the association between genotypic and phenotypic data, was performed with the following factorial model of covariance analysis with fixed factors utilizing SAS 9.1v software:

$$Y_{ijK} = \mu + b_1x_1 + b_2x_2 + b_3x_3 + \alpha_i + sex_j + e_{ijK}$$

where:

μ = constant common to all observations (overall mean);

x_1 = weight of refrigerated carcass after 72 hours of refrigeration (covariate);

x_2 = date of slaughtering (covariate);

x_3 = age of the pig at slaughtering (covariate);

α_i = fixed effect common to all observation relative to i^{th} genotype (i = 1,2,3)/haplotype (i=1, 2, 3, 4, 5, 6);

sex_j = fixed effect common to all observations related to j^{th} sex (j = 1,2);

e_{ijK} = random error.

III – Results and discussion

From statistical elaboration it emerged a significant effect of some *loci* on carcass fatness. *H-FABP*. The comparison among the considered haplotypes highlighted that the pig with HHDdAA or HHDdAa haplotype, when compared with a subject with HHddAA or HHddAa or HHddaa haplotype, gives a higher adiposity estimated through any detected parameters (Table 2). In particular, the difference of: (i) back fat thickness can vary from about 9 (HHDdAa *vs* HHddAa; P=0.020) to about 17 mm (HHDdAA *vs* HHddAA; P=0.002); (ii) total fat cuts (without belly) can range from 6.5 (HHDdAA *vs* HHddAa; P=0.075) to about 16 kg (HHDdAa *vs* HHddaa; P= 0.002); (iii) of separable cuts can vary from about 6.0 (HHDdAA *vs* HHddAa; P=0.073) to 13.5 kg (HHDdAa *vs* HHddAA; P=0.0001).

Table 2. Some parameters of fatness: significant comparisons between 'H-FABP' haplotypes

Haplotype		Parameter											
		Backfat thickness, mm						'Fat cuts', kg				'Total separable fat', kg	
Reference (I)	Comparison (J)	'Th1'		'Th15'		'L6-S'		'Total'		'Total without Jowl'			
		Δ (I-J)	P-value	Δ (I-J)	P-value	Δ (I-J)	P-value	Δ (I-J)	P-value	Δ (I-J)	P-value	Δ (I-J)	P-value
HHDdAA	HHddAA	12.443	0.031	15.113	0.004	16.949	0.002	8.047	0.028	8.197	0.042	10.542	0.005
	HHddAa	10.958	0.035	11.934	0.011	13.080	0.006	6.565	0.048	6.493	0.075	5.987	0.073
	HHddaa	15.412	0.043	15.277	0.027	16.563	0.017	5.154	0,287	13.485	0.013	6.677	0.173
HHDdAa	HHddAA	11.321	0.031	11.640	0.015	13.332	0.006	9.784	0.004	10.538	0.004	13.490	0.0001
	HHddAa	9.836	0.022	8.461	0.029	9.463	0.016	8.302	0.003	8.835	0.004	8.935	0.002
	HHddaa	14.290	0.043	11.804	0.063	12.946	0.043	6.890	0.126	15.827	0.002	9.625	0.036

The biological and operative importance to consider the effect of a haplotype on a quali-quantitative trait has been widely debated (Matassino *et al.*,1993; Zullo *et al.*, 1994). Indeed, according to these authors, the global genotype has a semantic value for marker assisted selection (MAS). Within H-FABP, it was believed convenient to individuate a possible effect of genotype at single *locus* on adiposity using single nucleotide polymorphism. The effect was significant only for D/d polymorphism: the subject with dd genotype gave a significant lower adiposity in comparison with heterozygote for the majority of the parameters considered, with a decrease equal to: (i) about 10 mm for back fat thickness (P= 0.003 at Th1 region; P= 0.001 at Th15 and L6-S regions); (ii) about 8 kg for total fat cuts and total separable fat (P<0.001).

FASN. The effect was significant on the weight of belly fat cut (P=0.051) and on that of separable fat from jowl (P=0.048). It is interesting to observe that the subject with TT genotype, in comparison with that with CC or CT genotype, gives a significant (P≤0.05): (i) lower weight of belly cut [the difference is equal to 0.895 kg and 1.072 kg for CC *vs* TT and CT *vs* TT comparisons, respectively]; (ii) higher weight of separable fat from jowl (difference equal to - 0.439 kg and - 0.454 for CC *vs* TT and CT *vs* TT comparisons, respectively).

DECR1. The effect of this gene was near to critical limit of significance (P= 0.072) on jowl; from the comparison between genotypes it emerges that the pig with CC genotype gives a lighter belly than the subject with CG or GG genotype; the comparison was significant (P= 0.022) for CC *vs* CG with a difference of -0.902 kg and tendentially significant (P=0.135) for CC *vs* GG with a difference of - 0.608 kg.

SCD. This gene tended to influence the weight of separable fat from coppa adipose cut (P= 0.148), back fat (P= 0.144) and total separable fat (P= 0.132).

MC4R. A tendency to significance (P= 0.179) was evidenced for separable fat from jowl.

IV –Conclusions

In the limits of the observation field, the results suggest that H-FABP haplotype significantly affects the parameters used for the estimation of carcass adiposity, determining differences among subjects with different haplotypes. The pig with HHddAA would be the less physiological obese for the total of adipose cuts without jowl and of separable fat. FASN locus would cause a differential fat deposition probably associated to the effect of TT genotype showing a variable lipogenetic aptitude in different anatomical regions of the body. For DECR1 locus, CC genotype would influence the weight of belly fat. Further investigations are ongoing for an operative utilisation of the H-FABP haplotype and FASN and DECR1 genes as molecular markers (candidates) in proper MAS plans.

Acknowledgements

The authors wish to acknowledge the financial support received from the Mipaaf. The authors want to thank dressed pork factory 'Di Maria' Cese Alte – Circello (BN).

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Effect of feeding and rearing system on growth performance of Sarda breed pig: Preliminary study

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Abstract. Autochthonous Sarda pigs are usually bred in mountainous areas of Sardinia where they use mainly feeding resources in shrublands and forests. The aim of the study was to evaluate the effect of different feeding systems on some performances of Sarda pigs. Twelve, castrated male pigs, homogeneous for body weight (98.4±13.8 kg) and age (15 months) were randomly assigned to the following three feeding systems: A, reared en *plein-air* system in the woods and fed *ad libitum* with a commercial concentrate; B, reared en *plein-air* in the woods and fed with ground barley (1.8 kg/head/day), with an automatic feeder; C, fed at pasture in the woods and receiving 500 g/head/day of barley grain. Morphological measures and weight were recorded fortnightly until slaughtering (22 months of age). Preliminary results showed differences between feeding systems, with best performance in group A: weight and fat thickness were higher (294 kg and 84.5 mm respectively) than group B (170.7 kg and 29.7 mm) and group C (202.0 kg and 39.7 mm). Therefore extensive feeding systems is a right compromise between good performances and farming profit.

Keywords. Sarda breed pig – Growth performance – Feeding system – Rearing system.

Etudes préliminaires sur l'influence de l'élevage et de l'alimentation sur l'augmentation pondérale et biométrique des porcs de race Sarde

Résumé. *Le porc autochtone de race Sarde, traditionnellement élevé dans les régions montagneuses de Sardaigne, se nourrit principalement des ressources du sous-bois. L'objectif de ce travail était d'étudier l'influence des divers systèmes d'alimentation sur les performances productives de ces porcs. Douze mâles castrés, de poids (98,4±13,8 kg) et d'âge (15 mois) comparables, ont été soumis à trois régimes alimentaires différents: A, élevés en plein air dans un bois et alimentés* ad libitum *avec un aliment commercial; B, élevés dans le même système et alimentés avec de la farine d'orge (1,8 kg/tête/jour) au moyen d'un distributeur automatique; C, maintenus au pâturage dans un bois et complémentés avec de l'orge en grain (500 g/tête/jour). Les mesures morphologiques et le poids de chacun de ces animaux ont été relevés tous les 15 jours jusqu'à leur abattage à 22 mois. Les résultats préliminaires ont montré des différences significatives (P<0,05) entre ces systèmes d'alimentation avec de meilleures performances pour le groupe A: le poids vif et l'épaisseur de gras des porcs de ce groupe (respectivement 294,0 kg et 84,5 mm) étaient supérieurs à ceux des groupes B (170,7 kg et 29,7 mm) et C (202,0 kg et 39,7 mm). Toutefois, le système d'alimentation extensive représente le meilleur compromis entre performances de production et profit de l'éleveur.*

Mots-clés. *Porc de race Sarde – Gain de poids – Système d'alimentation – Système d'élevage.*

I – Introduction

The increased industrialization of agriculture and the greater market demand led the pig farmers to replace the "autochthonous breeds pig", less productive but well adapted to the environment, with the "cosmopolitan breeds pig". This trend started in Italy, since 1872 (Mascheroni, 1927), and caused the gradual replacement of the autochthonous breed pigs.

Among the 21 Italian breed pigs, recognized until the last century, nowadays only 6 survived (Franci *et al.*, 2007). Despite ancient origins of Sarda pig (Porcu, 2006), the study about the local breed officially recognized (Ministerial Decree No. 21664 of 08/06/2006) has been started recently (Porcu *et al.*, 2007).

The exploitation of the autochthonous species can help the biodiversity safeguard and the fight against the depopulation of the marginal areas (Porcu, 2008). Indeed, in all rural societies, domestic pig breeding has played always an important role because represented, and represent still now, a source of food always available thanks to the spawning characteristics (high fertility and frequency of parts). *Sarda* pig has lived for centuries completely free in the Sardinia mountains showing a great ability to utilize poor food such as spontaneous fruits of woods and only occasionally was supplemented with flour or grains. Such as autochthonous pig (Zumbo *et al*., 2003), Sarda breed is characterized by a strong resistance to illness, a zoo-technical adaptability to different climatic conditions as well as the ability to procure food thanks to its strong inclination to grazing and its high rusticity.

Several authors (Cetti, 1774; Bonadonna, 1960; Porcu, 2004) described the Sarda breed pig as a small animal size that hardly could achieve heavy weight.

However Cetti (1774) stated that in some areas of Sardinia some pigs could reach greater size (over 200 kg). So is well-known that the productive and growth performances are influenced both by genetic factors and different environmental conditionings. Among these, the feeding and rearing systems should be considered particularly.

The aim of the study was to evaluate the effect of different breeding and feeding systems on growth performances of Sarda pigs.

II – Materials and methods

The study, lasted seven months (July 2009-February 2010) and was conducted at the experimental farm of AGRIS agency in Foresta Burgos, (Illorai, Sassari; 44° 69 lat. North and 4° 95 long. East). The experimental site, is characterized by a forest area of 40 ha divided in lots of various sizes and with altitude ranging from 830 to 930 m. Twelve Sarda breed castrated male pigs coming from the AGRIS agency (DIRPA), homogeneous for genetic, weight (98.4 ± 13.8 kg) and age (15 months) were randomly assigned to three experimental groups (A, B and C) that differed in the feeding systems: A, reared en *plein-air* system in a wood area of 4500 m^2 and supplemented with a commercial concentrate (*ad libitum)*; B, reared en *plein-air* system in a wood area of 6000 m^2 and supplemented with ground barley (1.8 kg/head/day), through an automatic feeder; C, fed at pasture in a wood area of approximately 20 ha and supplemented with barley grain (500 g/head/day). Chemical composition of principal feedstuff used and main fruits of woody species fed during the experiment was measured (Table 1).

Table 1. Chemical composition of feed (% dry matter)

	Moisture	Ash	Crude protein	Ether extract	Crude fiber
Quercus pubenscens	54.50	2.47	5.76	2.31	14.83
Quercus ilex	56.91	2.23	4.82	1.70	16.88
Pyrus amygdaliformis	63.62	2.47	3.35	1.1	22.68
Commercial concentrate	14.04	7.69	16.79	2.8	4.91
Ground barley	12.04	2.67	9.11	1.94	7.36

On each animal, fortnightly morphological measurements were determined: live weight, withers and rump height, chest length, chest depth, chest width, rump width and subcutaneous fat thickness at the lumbar level (by RENCO LEAN-Meter instruments). The animals were slaughtered 22 months old.

Data of live weight and morphological parameters were tested by GLM procedure (SAS, 2001) using feeding system as fixed effects. Average daily gains (ADG) were tested by MIXED procedure (SAS, 2001) using feeding system, measurement period and their interaction as fixed effects and animal as random effect.

III – Results and discussion

As expected, pigs fed *ad libitum* with commercial concentrate (group A) have grown faster than those fed in controlled condition (Fig. 1, Table 2) and between these two groups, higher ADG has been observed in C than B group (P<0.001).

Table 2. Effect of feeding regimen (TR), period (NC) and interaction on average daily gain (AMG) of Sarda pigs submitted to different feeding systems (ls means ± S.E.)

	N	Groups			P<		
		A	B	C	TR	NC	TR*NC
ADG (kg/head/day)	12	0.910±0.03 a	0.351±0.03 c	0.469±0.03 b	0.001	0.001	0.001

a, b values with different letters are different (P<0.05).

Preliminary results (Table 2) showed differences between feeding systems, with best performance in group A: weight and fat thickness were higher in group A than group B and group C (Table 2). This is probably due to the higher energetic availability in the A group. Also the fat thickness was higher (P<0,01) in group A than group C; this result is in agreement with data of Pugliese *et al.* (2003) on Nero Siciliano pigs reared indoor. The lower backfat thickness found in groups B and C should be considered in relationships with the lower growth rate observed in these groups. Pugliese *et al.* (2003) reported that a slower growth rate usually favours muscle deposition with respect to fat, resulting in leaner carcasses.

Table 3. Live weight and morphological measurements of Sarda breed submitted to different feeding systems (ls means ± S.E.)

	N	Groups			
		A	B	C	P<
Live weight (kg)	12	294.0±12.0 a	170.7±12.0 b	202.0±12.0 b	0.001
Withers height (cm)	12	89.7±1.6 a	82.0±1.6 b	85.7±1.6 b	0.020
Rump height "	12	91.0±1.5 a	83.5±1.5 c	88.0±1.5 b	0.020
Chest girth "	12	172.2±3.0 a	129.7±3.0 c	139.2±3.0 b	0.001
Chest lenght "	12	135.0±4.1 a	116.5±4.1 b	125.0±4.1 b	0.030
Chest depth "	12	59.0±1.2 a	45.7±1.2 b	49.5±1.2 b	0.001
Chest width "	12	46.0±0.7 a	35.7±0.7 b	37.0±0.7 b	0.001
Rump width "	12	43.7±0.9 a	31.5±0.9 c	34.5±0.9 b	0.001
Fat thickness (mm)	10	84.5±3.8 a	29.7±2.7 c	39.7±2.7 b	0.001

a, b values with different letters are different (P<0.05).

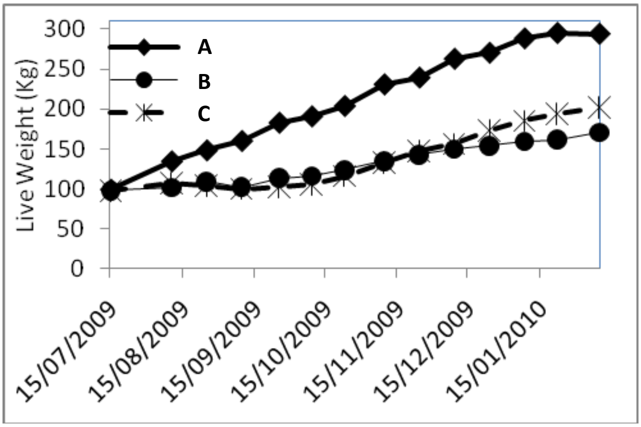

Fig. 1. Live weight trend (kg) of Sarda pigs submitted to different feeding systems (A, reared in plein-air system in the woods and fed *ad libitum* with a commercial concentrate; B, reared in plein-air in the woods and fed with ground barley (1.8 kg/h/d), with an automatic feeder; C, fed at pasture in the woods and receiving 500 g/h/d of barley grain).

IV – Conclusions

Data show that the performance *in vitam* were significantly influenced by the nutritional level as reported by Liotta *et al.* (2005). The extensive feeding system represents the best compromise between performance and farming profit. The preliminary results presented encourage to pursue research experiments to better understand which feeding and rearing conditions can allow good results by respecting the tradition and productive performance of Sarda breed pig.

Acknowledgements

The Authors want to thank S. Fois, G. Moro, A. Pireddu, L. Pitzolu, F. Ruiu, D. Tatti, G. Zoeddu, C. Manca, M. Delrio for their precious contribution.

The research was supported by Agriculture Assessorship of the Sardinia Region.

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Biometric and rheologic parameters and qualitative properties of meat from "Sarda" breed pigs: Preliminary results

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Abstract. The aim of the present study was to evaluate the impact of different breeding systems on the quality parameters of meat from Sarda pigs. Twelve male pigs, aging 15 months, were subdivided in three groups (A,B,C) homogeneous in number of subjects and mean weight (98.4±13.8 kg). Groups A and B were reared in *plein-air* and fed with commercial concentrate *ad libitum* and ground barley (1.8 kg/head/day) respectively. Group C was grazed on pasture and received a daily supplement of 500 g/head/day of barley grain. All the subjects were slaughtered after 7 months. A section between L2 to L5 was used to evaluate the lean, fat, rind, connective and bone yield. Samples of muscles *Longissimus dorsi* and *Psoas major* and inner fat were used to determinate colorimetric (CIELab) and rheologic (TPA test: 24h – 7days) parameters. Group A showed highest yields in fat (71.8%) and lowest in connective (1.2%) linked to lowest values in terms of hardness. Group B showed highest yields in lean (41.1%) and connective (2.6%) and lowest in fat (48.9%) linked to highest hardness and lowest springiness values. Group C showed intermediate values in lean (34.6%), fat (55.9%) and connective (1.5%) yields but the samples resulted more firm.

Keywords. Sarda breed pig – Colorimetric and rheologic parameters – Meat picking yield – Meat quality.

Paramètres biométriques, propriétés rhéologiques et qualitatives de la viande du porc autochtone de race Sarde: Résultats préliminaires

Résumé. *L'objectif de ce travail est l'évaluation de l'impact des techniques d'élevage sur les caractéristiques qualitatives de la viande du porc autochtone de race Sarde. Douze mâles (15 mois), divisés en trois groupes (A,B,C), de poids (98,4 ±13,8 kg) et d'âge comparables ont été soumis aux techniques d'élevage suivantes: en plein air, avec administration d'un aliment commercial* ad libitum *(A) ou d'une ration d'orge moulue (B); au pâturage dans un bois (C), avec une complémentation quotidienne de 500 g d'orge en grain. À l'abattage (22 mois) ont été mesurés les rendements en maigre, gras, couenne, conjonctif et os sur une section de la région L2-L. Les paramètres colorimétriques (CieLab) et rhéologiques (TPA test: 24h - 7 jours) ont été étudiés sur les échantillons des muscles* Longissimus dorsi *et* Psoas major. *Le groupe A montrait des rendements élevés en gras (71,8%) et faibles en conjonctif (1,2%), corrélés à des valeurs faibles de dureté. Le groupe B présentait des rendements élevés en maigre (41,1%) et conjonctif (2,6%) et faibles en gras (48,9%), qui s'accompagnaient de valeurs de dureté supérieures et de souplesse inférieures. Le groupe C présentait des valeurs intermédiaires en ce qui concerne les rendements en maigre (34,6%), gras (55,9%) et conjonctif (1,5%), mais les échantillons étaient plus compacts.*

Mots-clés. *Porc de race Sarde – Paramètres colorimétriques et rhéologiques – Rendement de carcasse – Qualité de la viande.*

I – Introduction

The preservation of the autochthonous breeds is an useful tool for the biodiversity safeguard in the mediterranean areas, where the economic sustainability and productivity of the ecosystem are an essential requirement for their subsistence. Moreover the autochthonous race breeding affects some important ecological, social and cultural aspects, such as safeguard of the regions, the rural community and their traditions.

Recently, initiatives in safeguard of autochthonous Sarda breed pigs were carried out in Sardinia, (Porcu *et al*., 2007), aimed at the valorization of meat and meat products. Consumers demand and appreciate these kind of products, where recognize genuineness, sustainability, as well as environment integration. On the other hand, due to the poor productive performance, the survival of this breed is strongly linked to the development of products.

Many researches showed that chemical composition and quality of meat of autochthonous breeds vary in relation to rearing system, age and weight at slaughtering (Lo Fiego *et al*., 2007), and in general, demonstrated non homogeneous characteristics (Gentry *et al*., 2004; Gonzales *et al*., 2007).

Few studies have been carried out about Sarda breed pigs (Porcu *et al*., 2010), that are usually reared by outdoors systems. Improve the knowledge on the quality of meat is an extremely important issue for its qualification, but you must also identify best breeding conditions to combining yields and quality.

The aim of the present study was to evaluate the impact of different breeding and feeding systems on some meat parameters of Sarda breed pigs.

II – Materials and methods

Twelve Sarda breed male pigs, with 98.4±13.8 kg of body weight, were subdivided into three groups (A,B,C), each including four animals. Groups A and B were reared in *plein-air* and allowed *ad libitum* access to commercial feed and rationed ground barley respectively. Group C was grazed on woody pasture and received a daily supplement of 500 g/head/day of barley grain. The pigs were weighed weekly during the experiment, which lasted seven months. After slaughtering, a section of the region included between the 2^{th} and 5^{th} lumbar vertebra (Campodoni *et al*., 1999; Porcu *et al*., 2007) was isolated and dissected into the major tissues (lean, fat, connective and bone). Samples of *Longissimus dorsi*, *Psoas major* and inner fat were analyzed for: (i) Colorimetric parameters: one hour after slaughtering, L*, a*, b*, values were determined, in triplicate, by a Chroma Meter Minolta CR400, standard illuminant C; and (ii) Texture analysis: both 24 hours and 7 days after slaughtering, two cylinders (1.5 x 2 cm) were obtained for each sample of the muscle. TPA test was performed using a Universal Testing Machine TAXT plus Texture Analyser (Stable Microsystems Ltd.) with the Texture Exponent software (Vs.2.0.0.7). A double compression cycle test was performed using an aluminum cylinder probe (P/75). A time of 0 s was allowed to elapse between the two compression cycles (bite). Force–time deformation curves were obtained with a 5 kg load cell applied at a cross-head speed of 1 mm/s. The following parameters were evaluated: *hardness* (g, H), maximum force required to compress the sample; *cohesiveness* (Co), extent to which the sample could be deformed prior to rupture; *springiness* (m), ability of the sample to recover its original form after deforming force was removed; *adhesiveness* (g x s), negative parameter which represent the area under the abscissa after the first compression. The results were analyzed using GLM procedure of SAS (2001).

III – Results and discussion

The evaluation of sample cuts (Table 1) showed the positive effects of different rearing and feeding systems on weight and fat percentage, being the mean values highest in Group A (6193.5±446 g and 71.8±2.5% respectively). Moreover samples from Group A showed lowest percentage in lean tissues (22.9%), connective (1.2%) and bone (4.1%).

Table 1. Tissue composition of the sample cut of Sarda breed pigs reared with different systems (ls means ± S.E.)

	N	Groups			
		A	B	C	P<
Sample cut weight (g)	12	6193±446 a	3396±446 b	4086±446 b	0.010
Lean (%)	12	22.9±1.4 c	41.1±1.4 a	34.6±1.4 b	0.001
Fat (%)	12	71.8±2.5 a	48.9±2.5 b	55.9±2.5 b	0.001
Connective (%)	12	1.2±0.3 b	2.6±0.3 a	1.5±0.3 b	0.050
Bone (%)	12	4.1±1.5	7.4±1.5	8.0±1.5	0.210

a, b means with different letters within row were significantly different (P<0.05).

The results of the colorimetric parameters are shown in Table 2. Low values of L* (range 38.7±1.6 → 51.9±2.2) were observed, especially in the *Psoas major*, where the minimum values were showed in samples from Group A. Meat from the Sarda breed pig appears darker and redder than commercial intensive reared breeds (Gentry *et al*., 2004), and other autochthonous breeds (Gonzales *et al*., 2007). Indeed the value of a*, positively correlated to meat quality, was very high, especially in muscle *Psoas major*, in comparison to other studies (Franci *et al*., 2004).

Table 2. Colorimetric parameters (TPA) in meat (muscles *Longissimus dorsi* and *Psoas major* and backfat) of Sarda breed pigs reared with different systems (ls means ± S.E.) at 24 h after slaughtering

	N	Groups			
		A	B	C	P<
Longissimus dorsi	12				
L*		48.3±2.2	50.1±2.2	51.9±2.2	0.53
a*		16.0±2.0	15.4±2.0	14.9±2.0	0.93
b*		9.9±1.2	10.0±1.2	10.1±1.2	0.99
Psoas major	12				
L*		38.7±1.6 b	41.3±1.6 b	46.0±1.6 a	0.02
a*		21.7±0.8	23.3±0.8	22.0±0.8	0.33
b*		9.3±0.6	10.9±0.6	11.7±0.6	0.07
Inner Fat	12				
L*		78.5±0.9	77.8±0.9	75.2±0.9	0.08
a*		1.4±0.2 c	3.6±0.2 a	2.8±0.2 b	0.001
b*		2.5±0.6	4.7±0.6	4.3±0.6	0.07

a, b means with different letters within row were significantly different (P<0.05).

The results of instrumental measurement of colorimetric parameters in inner fat showed a very high value of L* (mean value >75) and a significant difference between the groups regarding the a* value, that was higher in fat from samples from group B (3.6±0.2).

The results of the texture parameters in relation to the study groups, are shown in Table 3. Regarding the TPA test, significant differences between groups were not shown (p> 0.05) at 24 h nor 7 days after slaughtering. As expected, the hardness value decreased significantly with time in all samples. The springiness was higher in the samples from group A, and underwent a small increase during the maturation of meat at low temperatures. In samples from group C lowest values in terms of hardness were linked to the low percentage of connective. In group B highest hardness and lowest springiness values were correlated with the highest yields in lean

(41.1%), connective (2.6%) and lowest in fat (48.9%). The samples from group C resulted more firm and showed intermediate values in lean (34.6%), fat (55.9%) and connective (1.5%).

Table 3. Texture parameters (TPA) in meat (muscles *Longissimus dorsi* and *Psoas major*) of Sarda breed pigs reared with different systems (ls means ± S.E.) at 24 h after slaughtering

		Groups			
	N	A	B	C	P<
Longissimus dorsi					
Hardness (g)	12	11229±2764	16888±2764	14628±2764	0.38
Springness	12	0.7±0.05 a	0.5±0.05 b	0.6±0.05 b	0.05
Cohesiveness	12	0.5±0.05	0.4±0.05	0.5±0.05	0.65
Chewiness (g/cm^2)	12	3284±735	3031±735	3703±735	0.81
Adhesiveness (g x s')	12	-98.51±22	-66.5±22	-73.69±22	0.58
Psoas major					
Hardness (g)	12	13317±1375 a	9300±1375 b	8133±1375	0.05
Springness	12	0.5±0.04	0.5±0.04	0.6±0.04	0.39
Cohesiveness	12	0.4±0.05	0.4±0.05	0.4±0.05	0.98
Chewiness (g/cm^2)	12	3263±854	1829±854	1756±854	0.41
Adhesiveness (g x s')	12	-60.11±10	-32.28±10	-26.54±10	0.08

a, b means with different letters within row were significantly different (P<0.05).

IV – Conclusions

Although the preliminary results, Sarda breed pigs are incline to deposit subcutaneous fat when have greater food supply, as demonstrated in other autochthonous breeds. The data resulted comparable with other trials realized under similar conditions. However, further studies, aimed to identify the rearing and feeding systems suitable to obtain meat with high nutritional and technological values, will be carried out.

Acknowledgements

The Authors want to thank N. Lei, S. Pintus, and "La Genuina" Company (Ploaghe, SS, Italy) for their precious contribution. The research was supported by Agriculture Assessorship of the Sardinia Region

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Effects of feeding and rearing system on carcass characteristics of Sarda breed pig: Preliminary study

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Abstract. The effects of feeding and rearing systems on carcass characteristics of Sarda breed pigs were studied. Twelve castrated male pigs, homogeneous for body weight (98.4±13.8 kg) and age (15 months) were randomly assigned to three feeding systems. A e B groups, reared en plein-air system in the woods and fed *ad libitum* with commercial concentrate and fed ground barley (1.8 kg/head/day) respectively; C, fed at pasture in the woods and receiving 500 g/head/day of barley grain. At slaughtering (22 months) carcass weight, biometric measures, pH (at 45' and 24 h after slaughter) and backfat thickness located at the first (1T) and the last (UT) thoracic vertebra and at the top of the *middle gluteus* (GM) were recorded. The live weight (kg) value was 294.0 (A), 170.7 (B) and 202.0 (C). Results showed differences between feeding systems: slaughter yield at 1 h after slaughtering was higher in group A (82.04%) compared to B (78.03%) and C (77.97%); the slaughter yield at 24 h after slaughtering was 80.37% (A) *vs* 76.27% (B) *vs* 76.33% (C). Also the fat thickness was higher (P<0.01) in group A.

Keywords. Sarda breed pig – Feeding system – Rearing system – Slaughtering yield.

Effet du système d'alimentation et d'élevage sur les caractéristiques de la carcasse des porcs de race Sarde : Études préliminaires

Résumé. *Ce travail a consisté en l'étude des effets de l'alimentation et du système d'élevage sur le rendement à l'abattage des porcs de race Sarde. Douze mâles castrés, de poids (98,4±13,8 kg) et d'âge (15 mois) comparables, ont été divisés en trois groupes (A,B,C) et soumis à différentes techniques d'élevage: A et B, élevés en plein air, alimentés respectivement* ad libitum *avec un aliment commercial ou avec une ration de 1,8 kg/tête/jour de farine d'orge (B); C, au pâturage dans un bois avec une complémentation quotidienne de 500 g d'orge en grain. À l'abattage (22 mois) ont été mesurés: le rendement à chaud et à froid de la carcasse, les mesures biométriques, pH à 45' et à 24 h après abattage) et l'épaisseur du gras au niveau de la première et de la dernière vertèbre thoracique et du muscle* Gluteus *moyen. Le poids vif (kg) était de 293,4±7,2; 171,1 ±7,2 et 202,0±7,2 pour les groupes A, B et C. L'analyse des résultats a montré des différences entre les divers systèmes d'alimentation: le groupe A présentait un rendement à l'abattage supérieur, aussi bien à 1 h [82,04% vs 78,03% (B) et 77,97% (C)], qu'à 24 h [(80,37% vs 76,27% (B) et 76,33% (C)]. L'épaisseur de gras dorsal des carcasses du groupe A était aussi nettement supérieure (P<0,01).*

Mots-clés. *Porc de race Sarde - Système d'alimentation - Système d'élevage - Rendement à l'abattage.*

I – Introduction

The biodiversity safeguard and the fight against the depopulation of the marginal areas is given also by the exploitation of the autochthonous species (Porcu, 2008). Indeed, the use of autochthonous breed pig, some of which are living in absolute freedom, provide high quality production and increased the rate of self-provision of the meat market, limiting importation. Moreover they could allow a wider and more rational exploitation of marginal areas. The autochthonous breeds are the perfect balance between the animal and the environment in which they live increasing the value of the local traditions and diversifying the production

(Goracci, 2008). Sarda pig inserts in this context very well (Porcu *et al*., 2010).

Available data on productive performances and pork quality about free-range or confinement reared pigs vary widely vary: several factors could affect the results such as climatic and environmental conditions, genotypes etc. (Filetti *et al.,* 2003). Among these, in particular the nutritional level and qualitative aspects of the food should be considered. Also, the growth of the animal and its productive performance (chemical composition and meat quality) vary in function of rearing system, age and weight at slaughtering. Only recently few researches have been conducted about Sarda breed pig on their growth (Porcu *et al*., 2010). Establishing the performances of local breeds may involve both consistent differences between rearing environment and interactions between management and breed characteristics (Pugliese *et al*., 2003).

The aim of the study was to evaluate the effect of different breeding and feeding systems on carcass characteristics of Sarda breed pigs.

II – Materials and methods

The study lasted seven months (July 2009-February 2010) and was conducted at the experimental farm of AGRIS agency in Foresta Burgos (Illorai, Sassari; 44°69 lat. North and 4°95 long. East). The experimental site is characterized by a forest area of 40 ha divided in lots of various sizes and with altitude ranging from 830 to 930 m. Twelve Sarda breed pigs castrated male coming from the AGRIS agency (DIRPA), homogeneous for genetic, weight (98.4 ± 13.8 kg) , age (15 months) were randomly assigned to three experimental groups (A, B and C) that differed in the feeding systems: A, reared en *plein-air* system in a wood area of 4500 m^2 and supplemented with a commercial concentrate (*ad libitum)*; B, reared en *plein-air* in a wood area of 6000 m^2 and supplemented with ground barley (1.8 kg/head/day), through an automatic feeder; C, fed at pasture in the wood area of approximately 20 ha and supplemented with barley grain (500 g/head/day). Just before the slaughtering (22 months of age) live weight of each animal was recorded, then the carcass weight, the white offals weight and red offals weight were detected. The pH values: after 45 minutes (pH_1), after 24 hrs (pH_u), *post mortem,* at the top of the *middle gluteus* (GM), at the first (1T) and the last (UT) thoracic vertebra were recorded by a pH-meter (pH600 EUTECH Instrument) equipped with penetration probe.

After 24 hours of refrigeration at 4°C for each subject: carcass weight,and from the right half-carcass, pH_u and the thickness of dorsal fat located at the top of the middle gluteus (GM), at the first (1T) and the last (UT) thoracic vertebra, carcass length, inner and outer chest depth (Tables 2 and 3) were determined.

III – Results and discussion

The data (Table 1) showed that slaughter yield was significantly (P<0,001) higher in group A (82.04%) than B (78.03%) and C (77.97%).The slaughter yield at 24 h after slaughtering was 80.37% (A) *vs* 76.27% (B) *vs* 76.33% (C). Carcass length and chest depth were not different (Table 1). The fat thickness (Table 2), was higher in group A than the other ones.

Mean values of pH were reported in Table 3. No differences (P>0.05) were observed in pH_1, where the overall values were considered as normal for the examined muscles (range 6.15±0.07 → 6.55±0.11). Significant differences (P<0.05) between mean values of pH_u were observed (range 5.64±0.05 → 5.90±0.04). The pH_u values were highest in samples from group A (> 5.82), while resulted lowest in samples from group B (5.64).

The carcass characteristics showed the positive effects of different rearing and feeding systems on weight and fat thickness as reported by various authors (Liotta *et* al., 2005; Pugliese *et* al., 2003) on Nero Siciliano pigs fed *ad libitum* and reared indoor.

Table 1. Carcass yield at 0 and 24 h after slaughtering (ls means ± S.E.)

		Groups			
	N	A	B	C	P<
Live weight (kg)	12	294.0±12 a	170.7±12.0 b	202.0±12.0 b	0.001
Carcass weight 0 hr	12	241.2±9.7 a	133.2±9.7 b	157.5±9.7 b	0.001
Carcass weight 24 hr	12	236.3±9.4 a	130.2±9.4 b	154.2±9.4 b	0.001
White offals weight (kg)	12	13.6±1.2	17.1±1.2	14.6±1.2	0.18
Red offals weight (kg)	12	5.4±0.1 a	3.7±0.1 c	4.5±0.1 b	0.001
Carcass length (cm)	12	115.0±2.4	106.1±2.4	110.4±2.4	0.08
Chest depth (cm)					
Inner	12	25.6±1.1	23.4±1.1	27.1±1.1	0.12
Outer	12	29.1±1.5	26.9±1.5	28.9±1.5	0.51

a, b means with different letters within row were significantly different (P<0.05).

Table 2. Backfat thickness (mm) of Sarda breed pigs (ls means ± S.E.)

		Groups			
	N	A	B	C	P<
Fat thickness 1T					
Total	12	102±2.4 a	47±2.4 c	58±2.4 b	0.001
Inner	12	71±2.5 a	31±2.5 b	35.7±2.5 b	0.001
Fat thickness UT	12				
Total	12	108±3.1 a	49±3.1 b	56±3.1 b	0.001
Inner	12	79±4.3 a	32±4.3 b	41±4.3 b	0.001
Fat thickness GM					
Total	12	98±5.5 a	39±5.5 c	57±5.5 b	0.001
Inner	12	51±4.8 a	20±4.8 b	27±4.8 b	0.01

a, b means with different letters within row were significantly different (P<0.05).

Table 3. pH values of Sarda pig meat 45’ and 24 h after slaughtering (ls means ± S.E.)

		Groups			
	N	A	B	C	P<
pH_1 1T	12	6.28±0.07	6.24±0.07	6.15±0.07	0.44
pH_1 UT	12	6.29±0.07	6.21±0.07	6.19±0.07	0.30
pH_1 GM	12	6.55±0.11	6.47±0.11	6.44±0.11	0.70
pHu 1T	12	5.90±0.04	5.73±0.04	5.83±0.04	0.08
pHu UT	12	5.82±0.04 a	5.66±0.04 b	5.73±0.04 b	0.03
pHu GM	12	5.86±0.05 a	5.64±0.05 b	5.70±0.05 b	0.04

<a, b means with different letters within row were significantly different (P<0.05).

IV – Conclusions

Data show that feeding and rearing system affected carcass characteristics. The typical backfat content in this local breed can be an important issue. In-fact, in similar conditions, an high content on unsaturated fatty acids was found in adipose tissue of outdoor-pigs, particularly reared in wood (Pugliese *et al*., 2005; Cosentino *et al*., 2003). This aspect could be utilised to promote the quality of Sarda breed pig products. So the extensive feeding system represents the best compromise between performance and farming profit, as well as the valorization of local breed pigs can contribute to the economic and environmental sustainability of the traditional farming. However, being the data presented, the first scientific results obtained on the productive performance of Sarda breed pigs, further studies should be carried out in order to find the optimal rearing and feeding systems.

Acknowledgements

The Authors want to thank N. Lei, S. Pintus, G. Riu and "La Genuina" Company (Ploaghe, SS, Italy) for their precious contribution.

The research was supported by Agriculture Assessorship of the Sardinia Region.

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Characterization of dry-cured shoulders: Quality traits

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Abstract. The present study show some of the chemical and compositional characteristics of dry-cured shoulder from Iberian pigs fed with different diets and dry-cured shoulder produced with the rules of "Protected Designation of Origin Teruel ham". The existence of a quality rule makes the meat industries to follow different guidelines, from the selection of the genetic of the animal, food stuff and processing technology. As well, these rules establish different parameters that must be analyzed to avoid a possible fraud to the consumer. Not significant differences were found in the chemical parameters used by the quality standard, as fatty acids profile, in our samples. For this reason we must search other parameters to justify it. For that the use of products with characteristics very similar to the dry-cured ham which process time is shorter than that, like dry-cured shoulder, can help us to obtain good results which can be extrapolated. The aim of this study is to establish which structure and composition parameters of shoulders are more useful as quality indicators studying different dry-cured shoulders batches (white pigs *vs* Iberian pigs) depending on the genetic background of the animal and the type of feeding system during the final feeding phase.

Keywords. Shoulder – Protein – Lipid – Quality.

Une caractérisation des épaules séchées de porc : Des paramètres indicateurs de qualité

Résumé. *Dans le présent travail sont montrées certaines des caractéristiques concernant la composition des épaules séchées originaires de porcs Ibériques qui ont reçu divers types d'alimentation (aliments composés vs une alimentation naturelle) et des épaules séchées associées à l'"Appellation d'origine Teruel", qui sont élaborées conformément aux normes établies par leur appellation. La non existence de différences significatives pour quelques paramètres physiques et chimiques utilisés par la norme de qualité pour établir les caractéristiques de qualité que doivent présenter les produits, nous oblige à réaliser une nouvelle recherche de ceux-ci. Pour cela, l'utilisation de produits qui résistent à une élaboration plus rapide, tels que les épaules, peut servir à connaître le comportement d'autres produits comme le jambon d'élaboration la plus prolongée.*

Mots-clés. *Épaule – Protéine – Lipide – Qualité.*

I – Introduction

Dry-cured shoulders are products of high quality, however the most recognized product are dry-cured hams. Technological characteristics of dry-cured shoulders and sensorial qualities of the final product depend on many factors related to the production process. The effective efficiency related with shoulders production is directly connected with the genetic aptitudes and physiological characteristics (weigh, handing and genetic) where pigs raise.

The high proportion of bone and fat in the foreleg of the pig involves a lower yield and a complex consumption as consumers find it difficult to take advantage of the lean. Nevertheless, the high sensory quality, the nutritional value and the short ripening process make these products a very interesting issue of study for two main reasons. First, from a scientific point of view, the results can be useful as quality indicators of the raw material and the final product can be extrapolated to dry-cured ham which involves a longer ripening process. Secondly, an integral study of dry-cured shoulders according to its conformation by analysis of weight, measures, muscular topography and composition in order to develop a nutrition labeling (percentage of moisture, proteins, fat …), can be useful to establish a better utilization of these pieces and for the consumers to gain a greater knowledge of the quality of this product.

II – Materials and methods

The present study was carried out with three batches of Iberian dry-cured shoulders according to the type of feeding and rearing system during the finish fattening period (90 days prior to slaughter): (i) Country Valdesequera (n=9), (ii) Montanera Salamanca (n=10) and (iii) Normal intensive feeding (n=10), and (iv) another batch of dry-cured shoulder according to the "Protected Designation of Origin Teruel Ham", Teruel shoulders (n=20). Moisture, protein and sodium chloride percentage were determined using official methods (AOAC, 2000). Myoglobin content was evaluated using the method described by Horsney (1956). It was realized a TPA (texture profile) according to the method described by Bourne (1978) for a Universal texturometer TA-XT2i (Stable Micro Systems, Godaliming, UK). Intramuscular fat content (GIM) was extracted and quantify using a mixture of chloroform:methanol (2:1) according the method described by Folch *et al.* (1957). Fatty acids methyl esters (FAMEs) were prepared by acidic-trans-esterification in the presence of sulphuric acid (5% sulphuric acid in methanol) (Sandler and Karo, 1992). FAMEs were analyzed by gas chromatography using a Hewlett-Packard HP 5890A gas chromatograph, equipped with a flame ionisation detector (FID). Volatile compounds were extracted by using the solid-phase microextraction (SPME) and subsequently analyzed by gas chromatography coupled to mass spectrometry (GC/MS) (gas chromatograph Hewlett-Packard 5890 series II coupled to a mass selective detector Hewlett-Packard HP-5791 A) according to the method described by Jurado *et al*. (2007). Volatile compounds were tentatively identified by comparing their mass spectra with those reported in the Wiley and NIST libraries.

The results from the experiments were used as variables and analyzed using a multivariant analysis (SPSS, 1997) in order to compare physic-chemical parameters between batches. Statistical significance was predetermined at 0.05.

III – Results and discussion

Table 1 show high significant differences between batches, mainly the less weight and perimeter of the pieces of Montanera batch, it might be by the less initial weight of the pieces, because the yield of the novel pieces is in pure Iberian a 20% smaller than mixed Iberian, like intensive feed batch of this study. Teruel dry-cured shoulder was smaller in longitude and width because the age and size of the animals prior to slaughter was less than the Iberian pigs. Not significant differences were found between Montanera batch and extensive feeding batch (Country Valdesequera) in perimeter and width, probably because this shoulder came from of pure Iberian pigs (that's why these were longer for the same weight than the intensive feed batch).

Chemical composition is shown in Table 2. Not significant differences were found in the fat content between the four batches of the study. It might be because the batches feeding with feed used a prescription very similar than the acorn composition, according to the analysis of the fat composition from different batches. Montanera shoulders moisture was significantly smaller regarding to the other's shoulders, possibly because its morphologic sizes are smaller in this batch so the relation between surface and volume is higher. Myoglobin content showed significant differences being higher in Montanera and Country batches opposite the others two batches. These results are expected in animals raised in extensive. In others papers it had been related with quality attributes (juiciness, intensity of flavor and persistence of flavor), (Ventanas *et al*., 2007). Higher results of sodium chloride were found, (in Iberian ham are between 3.5%, and more than 4.5% are salty) in three batches of Iberian shoulder, in comparison with Teruel batch. This is because the three Iberian batches were processed in the same place and in the same conditions, different of Teruel shoulders. Finally, significant differences were found in protein percentage, being higher in Teruel batch.

Table 1. Morphological characteristics

	Country Valdesequera	Montanera Salamanca	Normal intensive feeding	Teruel shoulder	p
Weight (kg)	$5.4^{c} \pm 0.5$	$4.0^{a} \pm 0.5$	$5.5^{c} \pm 0.3$	$4.6^{b} \pm 0.4$	***
Lenght (cm)	$76.0^{c} \pm 3.6$	$71.4^{b} \pm 3.0$	$72.9^{b} \pm 0.5$	$62.6^{a} \pm 0.9$	***
Perimeter (cm)	$54.3^{ab} \pm 3.3$	$52.6^{a} \pm 1.4$	$56.0^{b} \pm 0.9$	$55.4^{b} \pm 1.2$	**
Width (cm)	$26.2^{a} \pm 3.6$	$26.0^{a} \pm 2.2$	$29.0^{b} \pm 3.2$	$23.9^{a} \pm 0.8$	***

Statistical significance: (***) p< 0.001; (**) p< 0.01; (*) p<0.05; (ns) not significant.

Table 2. Chemical composition

	Country Valdesequera	Montanera Salamanca	Normal intensive feeding	Teruel shoulders	p
% Moisture	$50.5^{b} \pm 3.2$	$46.8^{a} \pm 4.0$	$50.5^{b} \pm 3.5$	$50.5^{b} \pm 2.1$	*
% Chloride (NaCl) (DM)†	$11.7^{b} \pm 0.6$	$11.6^{b} \pm 1.7$	$11.6^{b} \pm 0.6$	$8.8^{a} \pm 1.1$	***
% Fat (DM)	13.6 ± 1.7	14.7 ± 3.5	14.1 ± 4.5	14.4 ± 2.2	ns
% Protein (DM)	$78.8^{ab} \pm 7.3$	$73.4^{a} \pm 7.1$	$84.1^{bc} \pm 8.9$	$87.5^{c} \pm 2.0$	***
Myoglobin (mg Mb/g m)	$6.6^{b} \pm 0.5$	$6.7^{b} \pm 0.4$	$3.9^{a} \pm 0.8$	$4.1^{a} \pm 0.6$	***

Statistical significance: (***) p< 0.001; (**) p< 0.01; (*) p<0.05; (ns) not significant.
†DM, dry matter.

Instrumental texture of Table 3 show the lower hardness of Teruel and Montanera batches in comparison with the others batches, because the higher content in fat make easier to broke the meat during the chewing, and the fewer gumminess and chewinness make higher the sensation of texture similar as "chewing gum".

Table 3. Instrumental texture

	Country Valdesequera	Montanera Salamanca	Normal intensive feeding	Teruel shoulder	p
Hardness	$3402.7^{b} \pm 724.8$	$2011.0^{a} \pm 616.6$	$3191.0^{b} \pm 1067.1$	$2037.4^{a} \pm 490.9$	***
Gumminess	$1521.6^{b} \pm 536.5$	$952.0^{a} \pm 334.1$	$1520.4^{b} \pm 532.8$	$930.1^{a} \pm 201.8$	***
Chewiness	$959.0^{b} \pm 379.1$	$523.4^{a} \pm 236.6$	$940.3^{b} \pm 368.7$	$571.0^{a} \pm 144.0$	***

Statistical significance: (***) p< 0.001; (**) p< 0.01; (*) p<0.05; (ns) not significant.

Table 4 show the total of the fatty acids presents in the intramuscular fat of the 4 batches analyzed. MUFA was significant higher in Montanera and Teruel batch. However the SFA was higher in Intensive feed batch; while in the Country batch the SFA value is similar than the Montanera and Teruel batches, not existing significant differences between them. To explain this, we need to know if the feed used is different for Country and Intensive feed batches. Other possible reasons could be because the fat content is higher in Country batch or because the genetic in this batch (pure Iberian) and the exercise make easier the desaturase activity. It is important the higher percentage of PUFA in Teruel shoulders, being similar than the Montanera shoulders. We expected these high results in comparison with the other Iberian batches of the study.

Table 4. Fatty acids profile

	Country Valdesequera	Montanera Salamanca	Normal intensive feeding	Teruel shoulders	p
SFA	$40.35^{ab} \pm 2.01$	$39.15^{a} \pm 0.98$	$42.53^{b} \pm 1.83$	$39.74^{a} \pm 2.84$	**
MUFA	$48.42^{a} \pm 1.92$	$50.01^{b} \pm 1.23$	$49.52^{ab} \pm 2.13$	$50.72^{b} \pm 1.22$	**
PUFA	$9.49^{ab} \pm 1.91$	$10.03^{b} \pm 1.94$	$8.07^{a} \pm 2.00$	$10.07^{b} \pm 0.74$	*
n-3	$0.85^{c} \pm 0.21$	$0.58^{b} \pm 0.11$	$0.39^{a} \pm 0.10$	0.61 ± 0.11	***
n-6	$8.14^{b} \pm 1.11$	$8.30^{b} \pm 1.54$	$6.52^{a} \pm 1.70$	$8.23^{b} \pm 0.58$	**
n-6/n-3	$8.75^{a} \pm 1.67$	$14.44^{b} \pm 0.97$	$17.56^{c} \pm 2.65$	$14.03^{b} \pm 0.52$	***

Statistical significance: (***) p< 0.001; (**) p< 0.01; (*) p<0.05; (ns) not significant.

Finally, were found significant differences in the content of n-3 between the four batches of the study, being higher in Country batch follow the others three batches. We expected this for Montanera batch because these pigs were fed with grass, but not for Teruel batch. We can explain this for the used feed; it can change a lot of parameters, like fatty acids or texture profile.

Table 5 shows some of the first results of the total volatile compounds (results in U.A.x10^{6}).

Table 5. Volatile compounds

	Country Valdesequera	Montanera Salamanca	Normal intensive feeding	Teruel shoulders	p
3-methyl-butanal	$9.0^{a} \pm 3.8$	$18.8^{a} \pm 10.7$	$36.3^{ab} \pm 30.2$	$62.6^{b} \pm 39.0$	***
2-methyl-butanal	$4.1^{a} \pm 2.3$	$8.1^{a} \pm 4.9$	$10.8^{a} \pm 7.1$	$24.5^{b} \pm 16.3$	***
Pentanal	$13.5^{a} \pm 6.7$	$13.8^{a} \pm 8.7$	$22.6^{ab} \pm 12.7$	$31.0^{b} \pm 17.2$	**
Hexanal	$106.0^{ab} \pm 45.6$	$113.6^{ab} \pm 55.7$	$143.9^{b} \pm 84.5$	$49.0^{a} \pm 40.4$	**
Heptanal	10.9 ± 5.2	11.0 ± 4.8	9.4 ± 4.4	8.6 ± 4.7	ns
Octanal	$6.7^{a} \pm 2.9$	$6.6^{a} \pm 2.5$	$8.8^{a} \pm 3.0$	$16.4^{b} \pm 12.0$	**
Nonanal	$10.9^{ab} \pm 6.0$	$12.2^{b} \pm 4.4$	$8.8^{ab} \pm 3.0$	$6.2^{a} \pm 4.0$	**
Decanal	$0.6^{a} \pm 0.3$	$0.4^{a} \pm 0.1$	$0.4^{a} \pm 0.1$	$1.4^{b} \pm 0.7$	***
2,3-butanedione	$3.3^{a} \pm 0.3$	$0.7^{a} \pm 0.3$	$1.1^{a} \pm 0.3$	$13.7^{b} \pm 6.0$	***
2-Heptanone	$29.0^{a} \pm 14.9$	$50.0^{b} \pm 32.6$	$26.1^{a} \pm 7.3$	$15.2^{a} \pm 9.1$	***
Dihydro 2(3H)-5-methyl-furanone	$0.8^{a} \pm 0.4$	$0.8^{a} \pm 0.1$	$0.6^{a} \pm 0.2$	$2.4^{b} \pm 1.4$	***
2-pentyl-furan	$2.4^{ab} \pm 0.2$	$5.6^{b} \pm 3.0$	$1.9^{a} \pm 0.5$	$9.4^{c} \pm 3.8$	***
Dihydro 2(3H)-5-ethyl-furanone	$3.4^{ab} \pm 1.8$	$2.4^{ab} \pm 0.9$	$1.7^{a} \pm 0.3$	$3.7^{b} \pm 2.0$	**
Dihydro 2(3H)-5-butyl-furanone	$0.9^{ab} \pm 0.4$	$0.5^{a} \pm 0.1$	$0.8^{ab} \pm 0.4$	$1.2^{b} \pm 0.4$	**

Statistical significance: (***) p< 0.001; (**) p< 0.01; (*) p<0.05; (ns) not significant.

The high number of volatile compounds detected, most of them waiting for identification, make us selected some of them because its interest. Is important the higher quantities in volatile compounds in Teruel shoulders probably because these shoulders had been under storage temperatures during the last stage of ripening, that improve the formation of this compounds like the case of the aldehydes. However, is amazing the hexanal behavior (rancid flavor), being higher in Iberian shoulders in comparison with Teruel shoulder. The high content in Iberian shoulders can be explained for its longer time of process in comparison with the Teruel shoulders, or for its higher concentration of precursors. In the case of ketones, we can emphasize the presence of 2-heptanone, presenting higher concentration in Montanera batch. One of the precursors of this ketone is linoleic acid that shows differences in content slightly higher in Montanera batch. However, this difference is not enough to explain the high difference in this compound between the Montanera batch with the rest of batches. This compound had been identified as odour-active compound showing aromatic notes like almonds or toast (Carrapiso *et al.,* 2002). At the end, 4 furans were identified and described as odour-active. These compounds were higher in Teruel shoulders and it could have an important paper in the aroma of the shoulders due to his presence in ham has related with the typical aroma to "meat" (Flores *et al.*, 1998). The fewer volatile content in the case of Iberian shoulders may be explained for the higher richness of aromatic notes for the presence of others volatile compounds which are analyzing actually.

IV –Conclusions

For the first time we provide a scientific data about shoulders quality, where the annual production is higher than 5 million of pieces, having a big economic weight in some communities in Spain.

With this study we can conclude that the fatty acids analysis can´t use only as indicator parameter about the quality of the product. In our case, we haven´t found any significant differences on the fat and the fatty acids proportion, existing the same tendency in the four batches. The same relationship had been found from the point of view of the genetic background and the animals feeding.

Acknowledgements

Authors thank to Juan García Cascos to provide us the samples for the study. Raquel Reina thanks Ana Antúnez for laboratory support and Julio Tapiador for his knowledge.

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Comparison between Cinta Senese and Mora Romagnola crossed with Large White pigs

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Abstract. The recovery of the "Prosciutto del Casentino", a typical product of the homonymous area located in the Arezzo province, led to the constitution, during 2007, of the "Consorzio del Prosciutto del Casentino". One of the main rules of the Consortium imposes the use of the cross between both Large White or Landrace with the Cinta Senese or Mora Romagnola autochthonous breeds. This first trial foreseen the comparison between two of the possible types of crossbreeding, in reason to the fact that the crossing between Cinta Senese and Large White is well investigated whereas information on the crossing between Mora Romagnola and Large White is lacking. Two different types of crossbreed have been compared for both *in vivo* and *post mortem* performances. Chemical-physical characteristics of raw meat and fat have been determined whereas the hams are at present during the seasoning period. MRxLW subjects produced fatter carcasses with higher thickness of subcutaneous fat than CSxLW (33.8 *vs* 22.9 mm). Sample joint of MRxLW showed higher fat and lower lean percentage than CSxLW (37.6 *vs* 25.2; 54.4 *vs* 64.7). As regards chemical-physical analyses it emerges that the meat of the CSxLW presented higher percentage of moisture (74.30 *vs* 72.71) and more water losses for pressure (100.05 *vs* 67.74 mm^2) than MRxLW.

Keywords. Ham – Cinta Senese – Mora Romagnola – Crossbreeding.

Comparaison entre Cinta Senese et Mora Romagnola croisés avec Large White

Résumé. *La récupération du Prosciutto Casentino, produit typique de la région homonyme dans la province d'Arezzo, a conduit à la création, en 2007, du Consortium de Prosciutto Casentino. Parmi les contraintes imposées par la réglementation se trouve l'utilisation de la première génération de croisements entre les races améliorées Large White et Landrace et les races indigènes Cinta Senese et Mora Romagnola. L'expérimentation a fourni une comparaison entre les deux types génétiques locaux, alors que, tandis que sont disponibles les données de croisement entre les Cinta Senese et Large White, les résultats de Mora Romagnola x Large White manquent. On a fait l'élevage des deux types de croisés, qui ont été comparés pour leurs performances* in vivo *et* post mortem*. Ensuite, on a déterminé les caractéristiques chimiques et physiques de la viande fraîche et de la graisse tandis que la maturation des jambons n'est pas encore terminée. Les animaux MRxLW ont produit des carcasses plus grasses avec une graisse sous-cutanée plus épaisse que CSxLW (33,8 mm* vs *22,9 mm) et des coupes plus riches en tissu adipeux et plus pauvres en maigre (37,6* vs *25,2, 54,4* vs *64,7). L'analyse physico-chimique montre que la viande de CSxLW a présenté un pourcentage plus élevé d'eau (74,30%* vs *72,71%) et une perte d'eau plus grande avec la méthode de la pression (100.05* vs *67,74 mm2).*

Mots-clés. *Jambon – Cinta Senese – Mora Romagnola – Croisement.*

I – Introduction

The use of the crossbreed between Cinta Senese and Large White pig in Tuscany has its origins early last century and provides the so-called "Bigio" or "Tramacchiato". This type of production was resumed last decade following the recovery of the Cinta Senese even if both the Technical Specifications of production for "Suino Cinto Toscano" and niche market, prefer the purebred. It remains that adequate test recently provided some guidance on the productivity of the cross of Large White with the Cinta Senese pig making possible a characterization, particularly in relation to the two parental breeds (Acciaioli *et al*., 2002; Franci *et al*., 2003, 2005). The characteristics of Mora Romagnola both as purebred or crossed are less known also because of its limited diffusion, which has hampered the interest of the research that only recently published results of tests and surveys on the breeding area (Fortina *et al*., 2005, 2006; Lo Fiego *et al*., 2007; Zambonelli and Bigi, 2006). The official inclusion of this breed in the Technical Specifications of a traditional production as the Casentino ham, determines the need to clarify the behaviour in relation to farming economy and quality of product. The aim of this study was therefore to assess qualitative differences between the two different crossbreeds Cinta Senese x Large White and Mora Romagnola x Large White.

II – Materials and methods

Eighteen pigs were used, 10 cross between Cinta Senese boars and Large White sows (CSxLW) and 8 cross between Mora Romagnola boars and Large White sows (MRxLW). The two genetic types, balanced by sex, were kept in two separate pens and fed mixtures *ad libitum*. During the period the subjects were weighed on a regular basis every two months. All the animals were slaughtered within two months starting from the heavier ones. At slaughter the carcasses were dissected in commercial cuts which were weighed to determine their percentage. On sample joint (loin from the 2nd to the 5th lumbar vertebrae) thickness of subcutaneous fat and area of the *Longissimus lumborum* were measured. Sample joint was dissected into: subcutaneous fat, divided into inner and outer layer, intermuscular fat, *Longissimus lumborum* and *Psoas major* muscles, other lean (muscle portions not identified) and bone. On *Longissimus lumborum* the following physical analyses were performed: (i) Color by Minolta colorimeter (Boccard *et al*. 1981); (ii) water holding capacity (Free water pressure by the method of Grau and Hamm (1952) modified; cooking loss (Boccard *et al*., 1981), drip loss; and (iii) shear force value by Warner-Bratzler instrument. On *Longissimus lumborum* and *Psoas major* the following chemical determinations were carried out: Moisture, ether extract, crude protein, Ash (AOAC, 1990). Data were processed by GLM procedure of SAS statistical package (2003) using the following models: Weight at slaughter, ADG and age $Y_{ijk} = \mu + R_i + S_j + \varepsilon_{ijk}$; carcass composition, sample joint composition and physical analyses of *L. lumborum* muscle $Y_{ijk} = \mu + + R_i + S_j + b^*(W_{ijk}) + \varepsilon_{ijk}$; Chemical analyses of *L. lumborum* and *Psoas major* muscles $Y_{ijkl} = \mu + R_i + S_j + T_k + b^*(W_{ijkl}) + \varepsilon_{ijkl}$; where R = Breed, S = Gender, T = Type of Muscle, W = weight of right side.

III – Results and discussion

Table 1 shows the in vivo performances of the two genetic types. The most evident result is the difference in slaughter weight, with the group CSxLW heavier at slaughter. This data could be affected by the difference in age between the two groups that is due to the choice of the breeder to slaughter the pig at the reaching of the visual maturity point. The most representative value was the ADG showing how the two groups behaved similarly during the trial period.

Table 1. Weight, ADG and slaughter age

Item	Genetic type		RSD
	CSxLW	MRxLW	
Live weight (kg)	179.3 a	155.4 b	21.6
ADG (kg)	0.352	0.378	0.06
Age (d)	504a	446b	14.2

As regard the carcass composition (Table 2), there was no difference in the dressing percentage but the results also showed that the CSxLW was basically leaner showing a greater proportion of ham and lesser proportion of kidney fat when compared to MRxLW.

Table 2. Composition of the right half side

Item	Genetic type		RSD
	CSxLW	MRxLW	
Dressing percentage	83.57	85.85	3.57
Loin with backfat	23.63 a	25.11 b	0.99
Ham (with feet)	32.17 a	28.94 b	0.93
Shoulder (with feet)	17.79	17.58	0.93
Belly with ribs	15.43	15.97	0.76
Jowl	3.30	3.36	0.46
Kidney fat	2.13 a	4.01 b	0.54
Head	5.55	5.03	0.38

Loin is greater in MRxLW only because this cut included backfat, according to the dissection protocol of the factory. The characteristic of MRxLW to depot more fat is clear also from the analyses of the sample joint (Table 3).

Table 3. Sample joint composition

Item	Genetic type		RSD
	CSxLW	MRxLW	
Sample joint (g)	2318.9	2482.1	3.57
Backfat thickness (mm)	22.98 a	33.84 b	4.14
Fat (%)	25.21 a	37.66 b	5.80
Backfat outer layer	12.19 a	15.75 b	2.05
Backfat inner layer	9.64 a	18.43 b	3.75
Intermuscular	3.70	4.27	1.16
Lean (%)	67.78 a	54.48 b	5.24
L. lumborum	40.12 a	33.80 b	3.25
Psoas major	14.98 a	10.59 b	2.03
Other lean	9.30	9.14	1.95
Bone (%)	10.06 a	8.02 b	1.37
Lean/bone	6.52	6.77	1.09

It is noteworthy that the thickness of subcutaneous fat is much higher in the group MRxLW. This parameter is confirmed by the incidence of subcutaneous fat, both as inner and outer layers. Intermuscular fat shows the same trend, even if not significant. Consequently to this behaviour of adipose tissue, the lean and bone components is more developed in CSxLW. Eliminating the masking effect of fat, however, the two genetic types showed similar lean/bone ratio.

The physical-chemical properties are reported in Table 4. The CSxLW showed moister meat, with lower content of protein and higher content of ash. Again, MRxLW showed higher fat content, even if not significant.

The moisture content also appear to affect the parameter of brightness (L*) which is higher in CSxLW subjects whereas the habit of the MRxLW animals to depot more fat didn't occur, showing how the influence of the genetic type is more evident in adipogenesis of the subcutaneous zone.

Table 4. Chemical analysis of lean

Item	Genetic type		RSD
	CSxLW	MRxLW	
Moisture (%)	74.30 a	72.71 b	1.01
Protein (%)	21.64 a	23.05 b	0.43
Ether extract (%)	2.35	2.91	1.18
Ash (%)	1.18 a	1.08 b	0.04

As regard the physical analyses of the meat (Table 5), CSxLW showed lower capacity of water retention only when measured as free water demonstrating that the three methods used for this analysis does not always agree among themselves as they are based on different physical principles of water removal.

Table 5. Physical analysis of *Longissimus lumborum*

Item	Genetic type		RSD
	CSxLW	MRxLW	
Color			
L*	52.01 a	48.37 b	2.16
a*	12.29	13.37	2.40
b*	5.10	4.90	1.04
Water holding capacity			
Drip loss (%)	6.83	7.18	1.89
Cooking loss (%)	24.20	24.88	4.06
Free water (mm2)	100.05 a	67.74 b	18.55
Shear force (kg)			
Wb fresh	10.08	9.29	2.53
Wb cooked	10.17	9.48	1.55

IV – Conclusions

The results showed that MRxLW crossbreed had greater tendency to depot fat. This trend is very evident in the subcutaneous fat and visceral fat. This could mean that this crossbreed is able to reach the slaughter age earlier, although at lower weights. On the contrary crossbreed including Cinta Senese breed favours lean cuts, especially the ham which is especially important from an economic point of view for the "Prosciutto del Casentino" production.

Acknowledgements

The authors wish to acknowledge the Agenzia Regionale Toscana per lo Sviluppo e l'Innovazione nel settore Agricolo e forestale (ARSIA), Provincia di Arezzo e Comunità Montana del Casentino for the financial support and Mr. Claudio Orlandi of the "Le Selve di Vallolmo" for the technical support.

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Vertical protein spot chains – proteomic indicators of proteolysis in dry-cured ham?

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Abstract. Proteomic profile of Slovenian "*Kraški pršut*" dry-cured ham matured for 14 months, was studied. Insoluble protein fraction was extracted from dry-cured *Biceps femoris* muscles. 2-dimensional SDS PAGE gels (24 samples in three technical repetitions) were made and the protein pattern analysed. Several distinctive protein spot patterns (*i.e.* protein spot chains containing spots that differed in molecular weight but not in isoelectric point) were observed. The patterns were highly repeatable between the technical repetitions. The subsequent identifications showed the same protein inside one spot chain. Differences in estimated molecular weight between the spots from the same chain indicate the protein degradation, however, it could not be confirmed by the mass spectrometry (lacking accuracy). For firmer confirmation of our hypothesis, a comparison of proteomic profile of hams in different processing phases is needed.

Keywords. Dry-cured ham – Proteolysis – Proteomic profile – Vertical spot chains.

Des chapelets verticaux de protéines : indicateurs protéomiques de la protéolyse dans le jambon sec ?

Résumé. *Le profil protéomique du jambon sec slovène "Kraški pršut" a été établi après 14 mois de séchage. La fraction de protéines insolubles a été extraite du mucle* Biceps femoris*. Les gels d'électrophorèse bidimensionelle SDS PAGE (24 échantillons en trois répétitions techniques) ont été réalisés ainsi que l'analyse d'image des gels. Plusieurs chaînes verticales de protéines distinctes (c'est-à-dire des spots protéiques différant en poids moléculaire, mais pas en point isoélectrique) ont été observées. L'apparition de ces chaînes était très reproductible entre les répétitions techniques. Les identifications ultérieures ont montré qu'il s'agit d'une même protéine à l'intérieur d'une chaîne. Les différences en poids moléculaire estimé entre protéines de la même chaîne indiquent leur dégradation progressive. Pour confirmer notre hypothèse, un suivi du profil protéomique des jambons au cours du séchage est nécessaire.*

Mots-clés. *Jambon sec – Protéolyse – Profil protéomique – Chapelets verticaux*.

I – Introduction

The proteolysis of muscle proteins by endogenous enzymes is one of the most important reactions that take place during dry-cured ham processing and is largely responsible for its sensory quality. The process itself begins already early *post mortem* with the breakdown of large cytoskeletal proteins (by calpains) and proceedes through degradation of myofibrilar proteins (mainly by cathepsins) and generation of great amount of small peptides and free aminoacids (by aminopeptidases), which may last for several months during the ham processing (Toldra and Flores, 1998). The degree of proteolysis can be assessed either directly by monitoring a degradation of several large proteins (in particular myofibrilar proteins) or indirectly by protein degradation products (shorter peptides, free amino acids and other amines, overall non-protein nitrogen). To evaluate degree of proteolysis chemical analysis of free amino acids or non protein nitrogen has thoroughly been used (Buscailhon and Monin, 1994a). One-dimensional protein electrophoresis has been used to follow up degradation of main muscle proteins (Toldra *et al.*, 1993; Tabilo *et al.*, 1999; Larrea *et al.*, 2006); however the method does not allow separation between different proteins of the same molecular weight. This is possible using a two-dimensional electrophoresis (2DE) which separates proteins according to their molecular weight and isoelectric point and which, coupled by mass spectrometry, enables identification of more than 1000 proteins in one gel (Gorg *et al*., 2000). This so called proteomic analysis represents a valuable tool for identification of molecular markers of food quality. Over the last years, several studies of proteomic research in meat science have been conducted (Hollung *et al.*, 2007). However the studies related to dry-cured ham are rare (Hortos *et al*., 2004; Di Luccia *et al.*, 2005; Sidhu *et al.*, 2005), moreover these studies are mainly preliminary and difficult to compare due to different methodology and approach. In our recent study (Škrlep *et al.*, 2010a) on dry-cured ham proteomic profile, we noticed several distinctive features on the dry-cured *Biceps femoris* gels, among which the vertical protein spot chains attracted the most of our attention. They could be an indication of progressive protein degradation, however, further characterisation would be needed to confirm that hypothesis, which was the aim of the present research.

II – Materials and methods

Material included in the present experiment originated from an extensive study on dry hams performed within EU project Truefood and experimental details are provided in our previous study (Škrlep *et al*., 2010b). The investigation included also the proteomic analysis of dry-cured *biceps femoris* (BF) muscle, for which a subsample of 24 hams was selected. Sample preparation, protein extraction and two dimensional electrophoresis (2DE) procedure is described in Škrlep *et al*. (2010a) and was performed according to the modified method developed at INRA (Theron *et al*., 2010). Shortly, insoluble protein fraction was extracted with repeated washing in low ionic strength buffer and loaded (1000 µg) on immobilised pH gradient strips for isoelectric focusing (70.000 Vh). For each sample three technical repetitions were made. SDS-PAGE was performed on 12.5% polyacrylamide gels. For the assessment of molecular weight (MW), protein MW marker #SM0431 (Fermentas Life Sciences, Glen Burnie, MD, USA) was applied prior to running second dimension. The gels were stained with Coomassie Brilliant Blue G250. The gel images were digitalized and spots automatically detected using ImageMaster 2D Platinum 6 software (GE Healthcare Bio-Sciences AB, Uppsala, Sweden). The comparison of the images from the present experiment to the images of fresh meat proteome from the available literature revealed some interesting differences, among which several vertical protein spot chains were the most distinctive (see Fig. 1). For the purpose of mass-spectrometry analysis, the spots of interest (spots from protein spot chains) were excised, destained, dehydrated, digested by trypsin and analysed (by peptide masss fingerprinting) using a Voyager DE Pro MALDITOF-MS (Applied Biosystems, Courtaboeuf, France) as previously described (Laville *et al.*, 2009). The obtained spectra were then compared to those from NCBInr databases susscrofa (20090623, 21575 seq) or mammalia (20090623, 1263710 seq) using Mascot Software (Matrix Sciences London, V2.2, home license),

III – Results and discussion

A representative 2DE gel image of insoluble muscle protein fraction of dry-cured BF muscle is shown on Fig. 1. In this article we focused on vertical chains of spots (also designated on Fig. 1). Such patterns could not be seen when compared to the corresponding regions of the gels reported for the fresh pig muscle (Morzel *et al*., 2004; Hwang *et al*., 2005; Laville *et al*., 2005). The chains of spots had almost the same isoelectrical point and different (app 0.5 – 1.0 kDa) estimated molecular weight (see Fig. 1). Some of the most distinctive spots from the chains were subsequently excised (n=16) and analysed by mass spectrometry. The results of the protein identification and peptide matching against the database records are shown in Fig. 2. In the first case (chain 1) all five spots (2932, 2357, 2404, 2423 and 2433) were identified as the same protein – myosin light chain (MLC1f). Three spots from the second chain (spots 2604, 2626 and 2653) were identified as another myosin light chain (HUMMLC2B) and one spot (2676) as fast skeletal myosin alkali light chain 1. For the remaining two analysed spot chains (spots 2457A, 2457, 2475, 2519 and 2558 in chain 3; spots 2576, 2617 and 2627 in chain 4) were again identified as the same protein, namely α-B-crystallin (chain 3) and myoglobin (chain 4).

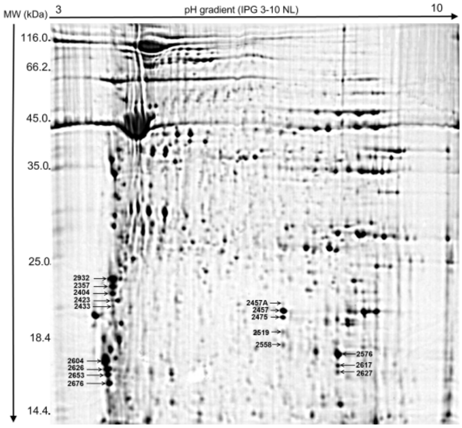

Fig. 1. Representative gel image of unsoluble protein fraction of dry-cured *Biceps femoris* muscle. Identified protein spots from vertical spot chains are denoted with arrows and reference numbers.

Since the pattern of vertical spot chains was repeatable between the technical repetitions (results not shown) showing clear separation of the spots with progressive decreasing of the estimated molecular weight, one explanation for that could be the gradual proteolytic degradation of the protein molecules. In agreement with our results several studies reported, that dry-cured ham proteins are prone to intensive hydrolysis during the processing period (Cordoba *et al*., 1994; 1993, Monin *et al*., 1997; Tabilo *et al*., 1999). More detailed studies, using one- and two- dimensional electrophoresis for monitoring the course of proteolysis (Di Luccia *et al*., 2005; Larrea *et al.*, 2006) reported notable degradation or even complete disappearance of several specific myifibrillar proteins (including myosin light chain) during the course of dry-cured ham processing, but direct comparison with the present study is not possible.

However, there are also several facts that speak against our hypothesis. The cleavage of protein spots is expected to cause the shift in isoelectric point, which is not the case for vertical spot chains. This could happen if the cleaved peptide (or aminoacids) were neutral or nonpolar. Furthermore, it could not be undoubtedly confirmed by the mass spectrometry results associated with protein database query that the spots were fragments (see Fig. 2), although the results of matching may indicate such conclusions (e.g. progressive decrease in sequence coverage and number of matched peptides), especially in the case of chains 1 (MLC) and 4 (myoglobin). It is also worth mentioning, that in the case of chain 1, the observed molecular MW of all five spots exceeds the MW of the theoretical match. The database query also did not give any myosin molecules, which would match our search closer in MW, making explanation even more difficult. However, Larrea *et al*. (2006) in a study supported by monodimensional SDS-PAGE, reported comparable myosin light chain of 24.75 kDa, which could match ours. It is worth noting, that we have not made any control gels on fresh ham muscle or muscle during different processing stages, which would be useful to clarify the origin of vertical spot chains.

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CHAIN 1.
Spot 2932 (gi|117660874, MLC1f)
  1 MAPKKDVKKP AAAAAPAPAP APAPAPAPAP PKEEKIDLSA IKIEFSKEQQ DEFKEAFLLF DRTGECKITL
 71 SQVGDVLRAL GTNPTNAEVK KVLGNPSNEE MNAKKIEFEQ FLPMLQAISN NKDQGSYEDF VEGLRVFDKE
141 GNGTVMGAEL RHVLATLGEK MKEEEVEALM AGQEDSNGCI NYEAFVKHIM SI
Spot 2357 (gi|117660874, MLC1f)
  1 MAPKKDVKKP AAAAAPAPAP APAPAPAPAP PKEEKIDLSA IKIEFSKEQQ DEFKEAFLLF DRTGECKITL
 71 SQVGDVLRAL GTNPTNAEVK KVLGNPSNEE MNAKKIEFEQ FLPMLQAISN NKDQGSYEDF VEGLRVFDKE
141 GNGTVMGAEL RHVLATLGEK MKEEEVEALM AGQEDSNGCI NYEAFVKHIM SI
Spot 2404 (gi|117660874, MLC1f)
  1 MAPKKDVKKP AAAAAPAPAP APAPAPAPAP PKEEKIDLSA IKIEFSKEQQ DEFKEAFLLF DRTGECKITL
 71 SQVGDVLRAL GTNPTNAEVK KVLGNPSNEE MNAKKIEFEQ FLPMLQAISN NKDQGSYEDF VEGLRVFDKE
141 GNGTVMGAEL RHVLATLGEK MKEEEVEALM AGQEDSNGCI NYEAFVKHIM SI
Spot 2423 (gi|117660874, MLC1f )
  1 MAPKKDVKKP AAAAAPAPAP APAPAPAPAP PKEEKIDLSA IKIEFSKEQQ DEFKEAFLLF DRTGECKITL
 71 SQVGDVLRAL GTNPTNAEVK KVLGNPSNEE MNAKKIEFEQ FLPMLQAISN NKDQGSYEDF VEGLRVFDKE
121 GNGTVMGAEL RHVLATLGEK MKEEEVEALM AGQEDSNGCI NYEAFVKHIM SI
Spot 2433 (gi|117660874, MLC1f)
  1 MAPKKDVKKP AAAAAPAPAP APAPAPAPAP PKEEKIDLSA IKIEFSKEQQ DEFKEAFLLF DRTGECKITL
 71 SQVGDVLRAL GTNPTNAEVK KVLGNPSNEE MNAKKIEFEQ FLPMLQAISN NKDQGSYEDF VEGLRVFDKE
141 GNGTVMGAEL RHVLATLGEK MKEEEVEALM AGQEDSNGCI NYEAFVKHIM SI

CHAIN 2.
Spot 2604 (gi|1176660856, HUMMLC2B)
  1 MAPKNAKRRA AAEGSSNVFS MFDQTQIQEF KEAFTVIDQN RDGIIDKEDL RDTFAAMGRL NVKNEELDAM
 71 MKEASGPINF TVFLTMFGEK LKGADPEDVI TGAFKVLDPE GKGTIKKHFL EELLTTQCDR FSQEEIKNMW
141 AAFPPDVGGN VDYKNICYVI THGDAKDQE
Spot 2626 (gi|1176660856, HUMMLC2B)
  1 MAPKNAKRRA AAEGSSNVFS MFDQTQIQEF KEAFTVIDQN RDGIIDKEDL RDTFAAMGRL NVKNEELDAM
 71 MKEASGPINF TVFLTMFGEK LKGADPEDVI TGAFKVLDPE GKGTIKKHFL EELLTTQCDR FSQEEIKNMW
141 AAFPPDVGGN VDYKNICYVI THGDAKDQE
Spot 2653 (gi|1176660856, HUMMLC2B)
  1 MAPKNAKRRA AAEGSSNVFS MFDQTQIQEF KEAFTVIDQN RDGIIDKEDL RDTFAAMGRL NVKNEELDAM
 71 MKEASGPINF TVFLTMFGEK LKGADPEDVI TGAFKVLDPE GKGTIKKHFL EELLTTQCDR FSQEEIKNMW
141 AAFPPDVGGN VDYKNICYVI THGDAKDQE
Spot 2676 (gi|1176660856, fast skeletal myosin alkali light chain 1)
  1 MSFSADQIAE FKEAFLLFDR TGECKITLSQ VGDVLRALGT NPTNAEVKKV LGNPSNEEMN AKKIEFEQFL
 71 PMLQAISNNK DQGSYEDFVE GLRVFDKEGN GTVMGAELRH VLATLGEKMK EEEVEALMAG QEDSNGCINY
141 EAFVKHIMSI

CHAIN 3.
Spot 2457A (gi|75063982, alpha-B-crystallin)
  1 MDIAIHHPWI RRPFFPFHSP SRLFDQFFGE HLLESDLFPA STSLSPFYFR PPSFLRAPSW IDTGLSEMRL
 71 EKDRFSVNLD VKHFSPEELK VKVLGDVIEV HGKHEERQDE HGFISREFHR KYRIPADVDP LTITSSLSSD
141 GVLTVNGPRR QASGPERTIP ITREEKPAVT AAPKK
Spot 2457 (gi|75063982, alpha-B-crystallin)
  1 MDIAIHHPWI RRPFFPFHSP SRLFDQFFGE HLLESDLFPA STSLSPFYFR PPSFLRAPSW IDTGLSEMRL
 71 EKDRFSVNLD VKHFSPEELK VKVLGDVIEV HGKHEERQDE HGFISREFHR KYRIPADVDP LTITSSLSSD
141 GVLTVNGPRR QASGPERTIP ITREEKPAVT AAPKK
Spot 2475 (gi|75063982, alpha-B-crystallin)
  1 MDIAIHHPWI RRPFFPFHSP SRLFDQFFGE HLLESDLFPA STSLSPFYFR PPSFLRAPSW IDTGLSEMRL
 71 EKDRFSVNLD VKHFSPEELK VKVLGDVIEV HGKHEERQDE HGFISREFHR KYRIPADVDP LTITSSLSSD
141 GVLTVNGPRR QASGPERTIP ITREEKPAVT AAPKK
Spot 2519 (gi|75063982, alpha-B-crystallin)
  1 MDIAIHHPWI RRPFFPFHSP SRLFDQFFGE HLLESDLFPA STSLSPFYFR PPSFLRAPSW IDTGLSEMRL
 71 EKDRFSVNLD VKHFSPEELK VKVLGDVIEV HGKHEERQDE HGFISREFHR KYRIPADVDP LTITSSLSSD
141 GVLTVNGPRR QASGPERTIP ITREEKPAVT AAPKK
Spot 2558 (gi|75063982, alpha-B-crystallin)
  1 MDIAIHHPWI RRPFFPFHSP SRLFDQFFGE HLLESDLFPA STSLSPFYFR PPSFLRAPSW IDTGLSEMRL
 71 EKDRFSVNLD VKHFSPEELK VKVLGDVIEV HGKHEERQDE HGFISREFHR KYRIPADVDP LTITSSLSSD
141 GVLTVNGPRR QASGPERTIP ITREEKPAVT AAPKK

CHAIN 4.
Spot 2576 (gi|47523546, myoglobin)
  1 MGLSDGEWQL VLNVWGKVEA DVAGHGQEVL IRLFKGHPET LEKFDKFKHL KSEDEMKASE DLKKHGNTVL
 71 TALGGILKKK GHHEAELTPL AQSHATKHKI PVKYLEFISE AIIQVLQSKH PGDFGADAQG AMSKALELFR
141 NDMAAKYKEL GFQG
Spot 2617 (gi|3660246, myoglobin)
  1 GLSDGEWQLV LNVWGKVEAD VAGHGQEVLI RLFKGHPETL EKFDKFKHLK SEDEMKASED LKKHGNTVLT
 61 ALGGILKKKG HHEAELTPLA QSHATKHKIP VKYLEFISEA IIQVLQSKHP GDFGADAQGA MSKALELFRN
141 DMAAKYKELG FQG
Spot 2627 (gi|47523546, myoglobin)
  1 MGLSDGEWQL VLNVWGKVEA DVAGHGQEVL IRLFKGHPET LEKFDKFKHL KSEDEMKASE DLKKHGNTVL
 71 TALGGILKKK GHHEAELTPL AQSHATKHKI PVKYLEFISE AIIQVLQSKH PGDFGADAQG AMSKALELFR
141 NDMAAKYKEL GFQG
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Fig. 2. Peptide matching against the database records in case of all four identified protein spot chains (matched sequences designated in bald and underlined).

IV – Conclusions

Although we could not undoubtedly prove that protein spot chains indicate proteolytic degradation of several dry-cured ham proteins, there is a strong indication towards our hypothesis. However, confirmation of our results is needed, by comparing the proteomic profiles of dry-cured ham in different processing stages.

Acknowledgments

The present study was financed by the Slovenian Research Agency (Project L4-9468) and the European Commission under the 6th Framework Programme for RTD (Project TRUEFOOD - "Traditional United Europe Food", contract n. FOOD-CT-2006-016264). The information in this document reflects only the author's views and the Community is not liable for any use that may be made of the information contained therein.

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Effect of production system and sex on different carcass traits of Iberian Pigs

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Abstract. Ninety *Retintos* Iberian pigs pure breed (45 males and 45 females) with similar age (~ 12 months old), live weight (90±5 kg) and the same genetic line (Line Valdesequera), were randomly selected and distributed into three groups (*Montanera*, *Recebo* and *Intensive*, n= 15 males and 15 females in each one) and slaughter at similar weight (150±10kg) for to evaluate the effect of production system and sex on some carcass traits. The length and width of rib cage, ham's perimeter, the thickness subcutaneous backfat (I, II and III levels) and the weight of carcass, loins, hams and forelegs were determined. The results showed that the production system had a significant effect on some carcass traits. So, the width of rib cage and the carcass's weight of pigs from *Montanera* were higher than the others systems. The thickness of subcutaneous backfat only shown differences in the measure most caudal, being higher in *Recebo* system and the weight of *Longissimus dorsii* (LD) was higher in *Intensive* system. On the other hand, the sex had effect on weight of forelegs (left and right), being higher in males in both pieces. In conclusion, yield cuts of the main commercial pieces (the hams) of the Iberian pig was not affected by the variables studied and males and females castrated only were different in the yield of forelegs.

Keywords. Pig – Ibéricos – Production systems – Carcass – Cutting.

Effet du système de production et du sexe sur différentes caractéristiques de la carcasse du porc Ibérique

Résumé. *Quatre-vingt-dix porcs de race pure Retintos Ibériques (45 mâles et 45 femelles) d'âge similaire (~ 12 mois), de poids vif (90 ± 5 kg) et de la même lignée génétique (Line Valdesequera), ont été choisis au hasard et répartis en trois groupes (Montanera, Recebo et intensif, n = 15 mâles et 15 femelles pour chacun) abattus à poids similaire (150 ± 10 kg) pour évaluer l'effet du système de production et du sexe sur certaines caractéristiques de la carcasse. La longueur et la largeur de la cage thoracique, le périmètre du jambon, l'épaisseur sous-cutanée du lard dorsal (niveaux I, II et III) et le poids de la carcasse, des longes, des jambons et des pattes ont été déterminés. Les résultats ont montré que le système de production a eu un effet significatif sur certaines caractéristiques de la carcasse. Ainsi, la largeur de la cage thoracique et le poids de la carcasse de porcs en provenance de Montanera étaient plus élevés que pour les autres systèmes. L'épaisseur sous-cutanée de lard dorsal ne fait apparaître des différences que pour la mesure la plus caudale, plus élevée dans le système Recebo, et le poids du* Longissimus dorsii *(LD), qui était plus élevé dans le système intensif. D'autre part, le sexe a eu un effet sur le poids des membres antérieurs (gauche et droit), plus élevé chez les mâles pour les deux pièces. En conclusion, les réductions de rendement des principales pièces commerciales (les jambons) du porc Ibérique n'ont pas été affectées par les variables étudiées et les mâles et femelles castrés étaient différents seulement pour le rendement des membres antérieurs.*

Mots-clés. *Porc Ibérique – Systèmes de production – Carcasse – Coupe.*

I – Introduction

The Iberian pig is the most important Mediterranean swine breed, both in population and economic importance (Serra *et al.*, 1998). Most Iberian pig is consumed as high-priced cured products. However, the consumption as fresh meat pork has recently increased in importance (Ramirez and Cava, 2005).

The main production system of Iberian pigs according to Spanish legislation (BOE, 2007) are *Montanera* (the typical free-range rearing system of the Iberian pig with a nutritional strategy based on acorns and grass), *Recebo* (free-range system with nutrition based on a combination of acorn, grass and concentrate), and *Intensive* (indoor system with nutrition based on concentrate).

The carcass is the first step in the process of production of meat. Currently, most commercial transactions in the meat market are made on the carcass. Quality and composition of the carcass depend on several factors among which include the feeding, the production system employed, sex or age. So, the main aim was to study the effect of the production system and sex of Iberian pigs on some traits of the carcass.

II – Materials and methods

Ninety Retintos Iberian pigs pure breed (45 castrated males and 45 females) with similar age (~ 12 months old), live weight (90±5 kg) and the same genetic line (Line Valdesequera), were randomly selected and distributed into three groups (n= 15 castrated males and 15 females in each one): *Montanera* (extensively reared and fed on acorns and grass), *Intensive* (intensively reared and fed on concentrate) and *Recebo* (extensively reared and fed on acorn and concentrate). Animals were slaughtered at 150±10 kg and ~15 months of age by electrical stunning and exsanguinations at a local slaughterhouse. After slaughtering, the carcasses were split longitudinally and weighed (carcass weight, kg). Carcass length was measured as the distance from the first rib to the pubic symphasis (lenght rib cage, cm) and carcass width was measured as drawing a straight line from the spine to the lower edge of the sternum (width rib cage, cm). On carcass, maximum perimeter ham (cm) was measured and also thickness subcutaneous backfat at three anatomical locations: the first rib (thickness subcutaneous backfat 1), the last rib (thickness subcutaneous backfat 2) and last lumbar vertebrae (thickness subcutaneous backfat 3). The left hand side of the carcass was divided into commercial cuts and commercial pieces weight were measured (kg): *Longissimus dorsii* (LD) muscle, left and right hams and left and right forelegs.

The effect of production system, sex and interaction of both variables was determined by analysis of variance, using the statistical software SPSS for Windows. If the effects studied was significant ($p<0.05$), then Tukey's test was used at the 5% levels to make comparisons between sample means.

III – Results and discussion

Results were shown in Table 1. No differences were observed between castrated males and females into the studied variables, except in the values of weight of the forelegs (left and right) which were higher ($p<0.01$) in castrated males. Mayoral *et al.* (1999) also found no difference between different sex on carcass length, LD's weight or subcutaneous fat thickness. Although, the slaughter weight was similar for all animals, the carcass of pigs from *Montanera* were higher ($p<0.001$) than the animals from others production systems. In previous studies with other rustic breed (Alentejo pig), Oliveira (2007) not found differences in carcass weight as affected by the production systems (indoor *vs* outdoor). Carcass measured indicate that length of rib cage and perimeter of ham were not affected by the production system; while the values of width rib cage were higher ($p<0.001$) in animals from *Intensive* system. Bridi *et al.* (1997) also not found differences in the length of rib cage affected by production system.

Thickness subcutaneous backfat were only affected by the production system in the last lumbar vertebrae (Thickness subcutaneous backfat 3) and the highest thickness was found in animals from *Recebo* while the least thickness in animals from *Intensive* systems. A contrary trend was observed by Chiofalo *et al*. (2007) in previous studies with Nero Siciliano pigs in the anatomical locations similar to our measure of thickness subcutaneous backfat 2. These differences could be due to both genetic and different production systems used.

Weight of commercial cuts (high value meat cuts) were not significantly affected by the production system except the *LD* muscle weight which was significantly greater (p<0.001) in the *Intensive* animals.

Table1. Effect of production system and sex on different carcass traits of Iberian pigs

	Production system			Sex		SEM	Significance	
	"Montanera" **n=30**	***"Intensive"*** **n=30**	***"Recebo"*** **n=30**	**Male n=45**	**Female n=45**		**Production system**	**Sex**
Lenght rib cage (cm)	80.83a	79.32a	78.8a	79.82	79.48	0.49	ns[1]	ns
Width rib cage (cm)	23.8a	25.81b	25.55b	24.82	25.29	0.19	***	ns
Perimeter ham (cm)	75.93a	74.65a	76.25a	77.00	74.21	0.62	ns	ns
Thickness subcutaneous backfat (cm)								
• first rib	8.97a	8.61a	8.8a	8.64	8.95	0.10	ns	ns
• last rib	6.65a	7.6a	7.03a	6.71	7.47	0.52	ns	ns
• last lumbar vert.	5.56ab	5.18a	5.84b	5.59	5.46	0.07	**	ns
Carcass weight (kg)	138.94b	131.04a	134.4a	136.40	133.18	0.79	***	ns
Longissimus dorsii muscle left weight (kg)	1.85a	2.1b	1.83a	1.90	1.95	0.02	***	ns
Ham (kg)								
• left weight	13.01a	12.97a	12.62a	13.14	12.59	0.17	ns	ns
• right weight	12.93a	12.89a	12.57a	13.05	12.54	0.16	ns	ns
Foreleg (kg)								
• left weight	9.07b	8.85a	8.84a	9.26	8.57	0.15	ns	**
• right weight	9.07b	8.84a	8.76a	9.25	8.53	0.14	ns	**

n: Number of samples. *SEM:* Standard error of the mean. ns: Not significant (*P*>0.05). Significant differences: ***: *P* ≤ 0.001; **: *P*≤0.01; *:*P*≤0.05).

IV – Conclusions

According to our results, castrated males and females showed a similar trend on thickness of subcutaneous backfat, yield of carcass and main commercial pieces (loin and hams), except in forelegs, which were heavier in castrated males than females. On the other hand, the production system affected to carcass and loin yield, being higher in *Montanera* and *Intensive*, repectively. No influences were observed on others commercial pieces yield, such as hams and forelegs. Furthermore, with of rib cage was lower in animals from *Montanera* system.

The descriptive statistics are presented as means (expressed as g water/kg fresh meat); values with the same letters (a, b, c) indicate homogeneous subsets for P=0.05 according to Tukey's HSD test.

Acknowledgements

The authors wish to acknowledge the financial support received from the Junta de Extremadura+FEDER (Project nº 564) and INIA+ Junta de Extremadura+FEDER (Projects nº RTA-2007-00093-00-00/JUN-08-563).

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Comparison of technological quality of meat and nutritional quality of backfat of Krškopolje pigs and commercial fatteners in Slovenia

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Abstract. Carcass traits, technological quality of *m. longissimus dorsi* and nutritional quality of back subcutaneous adipose tissue of autochthonous Krškopolje pig (KP) and commercial fatteners were compared. Commercial fatteners were classified according to lean meat content in fatty (51.3%), normal (57.9%) and meaty (64.0%) groups. In KP lean meat percentage was 47.8%. *M. Longissimus dorsi* of KP had the lowest pH_{24}, L* and h* and the highest a* values. The backfat in KP had the lowest a* and c*. The highest contents of IMF (1.96 and 1.94%) were observed in KP and fatty groups, respectively. Furthermore, the proportion of saturated fatty acid (SFA) in backfat did not vary between the groups. KP had the highest proportion of monounsaturated fatty acid (50.8%). Lower proportion of polyunsaturated fatty acid (PUFA) was also found in KP compared to the fatty group. Additionally, KP contained 5% less n-6 PUFA than the fatty group. The n-6/n-3 PUFA ratio of the meaty group was significantly higher than in KP. There were no differences in ratio of PUFA to SFA and atherogenic indices among the groups. In conclusion, KP had a better technological quality and nutritional quality of backfat compared to the fatty group of commercial pigs. It was very similar to the normal and meaty groups.

Keywords. Krškopolje pigs – Commercial fatteners – Technological meat quality – Nutritional quality.

Comparaison de la qualité technologique et nutritionnelle de la viande de gras du dos du porc Kraškopolje par rapport à ceux de porcs engraissés industriellement en Slovénie

Résumé. *La qualité de la carcasse, la qualité technologique du muscle Longissimus dorsi et la qualité nutritionnelle de la longe de tissu adipeux sous-cutané du cochon de race Kraškopolje (KP) ont été comparées à celles de porcs engraissés commercialement. La viande engraissée industriellement a été classée en trois groupes suivant la maigreur de la viande : grasse (51,3%), normale (57,9%) et charnue (viandeuse, 64%). Pour le groupe KP, le pourcentage de viande maigre représente 47,8%. Le muscle Longissimus dorsi provenant du groupe KP a les plus faibles valeurs de pH24, L* et h* mais aussi la plus haute valeur pour a*. Le gras du dos dans le groupe KP présente les valeurs a* et c* les plus basses. Les pourcentages plus élevés d'IMF ont été observés respectivement dans les groupes KP (1,96%) et viande grasse (1,94%). Enfin, la proportion d'acides gras saturés (SFA) contenus dans le gras du dos ne varie pas entre les groupes. Le groupe KP est celui qui contient la plus forte proportion d'acides gras mono-insaturés (50,8%). Enfin, il est à noter qu'une plus faible proportion d'acides gras poly-insaturés (PUFA) est retrouvée dans le groupe KP par rapport au groupe viande grasse. C'est le cas pour n-6 PUFA dont la quantité est plus basse de 5% dans le groupe KP que dans le groupe viande grasse. Le rapport n-6/n-3 PUFA est significativement plus élévé dans le groupe viande charnue que dans le groupe KP. Enfin, aucune différence n'a été mise en évidence pour le ratio PUFA/SFA et les indices athérogènes entre les différents groupes. En conclusion, la qualité technologique et nutritionnelle de la longe de tissu adipeux est meilleure dans le groupe KP que dans le groupe viande grasse des porcs élevés industriellement. Enfin, les résultats et les valeurs obtenues pour les groupes viande normale et charnu sont très similaires.*

Mots-clés. *Porc Kraškopolje – Engraisseurs industriels – Qualité technologique de la viande – Qualité nutritionnelle.*

I – Introduction

The Slovenian indigenous Krškopolje pig (KP) is usually under extensive rearing conditions.

Production traits of KP were studied in the past (Ferjan, 1969; Eiselt, 1971). Latter fewer researches were done. In the last decade Kastelic (2001) mentioned meat quality parameters of Krškopolje pigs. Furthermore, Čandek Potokar *et al.* (2003) have compared the carcass traits as well as the technological and sensorial quality of KP with its cross with a modern landrace line – LN 55. However, the composition of KP meat has not yet been investigated.

Technological quality is very important for meat processing. It is determined by technological traits such as pH value, electrical conductivity and colour. Firstly, the effect of pH value and electrical conductivity on technological quality was confirmed by Blendl *et al*. (1991). Further, meat colour, which can be effected by pH value and drip loss, often influences the consumer's choice of product (Ngapo *et al*., 2004). Technological quality of indigenous pig meat is interesting research topic because of its special meat characteristic.

The nutritional quality of meat has been paid considerable attention in research because of its implications for human health. The World Health Organisation (WHO, 2003) recommended an intake of 15-30% energy from fat, with less than 10% of this amount consisting of saturated fatty acids (SFA), 5-8% consisting of n-6 polyunsaturated fatty acids (PUFA) and 1-2% of n-3 PUFA. The nutritional recommendation for the n-6/n-3 PUFA ratio is less than 4:1 (Enser *et al*., 2001). The target ratio of polyunsaturated to saturated fatty acids (P/S) is 0.4 or above and the atherogenic index (AI) should be lower than 0.5 (Ulbricht and Southgate, 1991). To avoid consuming too much fat, people want to purchase lean meat. Therefore, reducing carcass fatness was one of the major breeding goals in farm animals for many years. However, it was likely to be accompanied by lower intramuscular fat levels (De Smet *et al*., 2004), and this had a negative influence on the sensory quality of meat. Dunn (1996) discussed that fattier pigs have more marbling which reflects better meat quality. This is one of the reasons for the better eating quality of meat of indigenous breeds compared to modern breeds.

Very little research has been done on the meat quality of the Slovenian indigenous breed Krškopolje pig. The present study compared the technological of *m. Longissimus dorsi* (LD) and nutritional quality of back subcutaneous fat tissue in Krškopolje pigs and commercial fatteners in Slovenia.

II – Material and methods

Ten KP originating from a small organic farm in the Pomurje region, were fed with organic feed in outdoor conditions. Commercial fatteners were fed with standard fattening feed mixture in a conventional indoor environment. On the slaughter line 43 commercial fatteners were randomly chosen. In order to compare the meat and fat quality, three groups of commercial fatteners were formed according to lean meat content.

Carcass traits for all animals were measured using the standard Slovenian on-line grading system at slaughter (EC, 2005). After slaughter the warm carcasses were weighted. The pH was obtained 24 hrs (pH_{24}) post mortem by pH meter Metter Toledo (MA130 Ion Meter) in LD and *m. semimembranosus* (SM). The electrical conductivity was measured with conductometer LF/PT-STAR (Matthäus) also in LD and SM 24 hrs post mortem ($COND_{24}$). Colour (L*, a*, b*) was measured in LD cut at the last rib 24 hrs post mortem by Minolta Chromameter CR300 (Minolta Camera Co., Osaka, Japan).

Samples of LD and back subcutaneous adipose tissue were taken 24 hrs after slaughter at the last rib. They were packed in vacuum bags and stored frozen at -21°C ± 1°C until chemical analyses. Intramuscular fat content (IMF) in muscle samples was determined by the Weibull-Stoldt method (AOAC, 1997). Fatty acids methyl esters (FAME-s) from samples were prepared using the Park and Goins method (1994). The results were expressed as a percentage of total fatty acids. The statistical model included group effect (G_i) and adjustment for carcass weight. Analysis was carried out using the GLM procedure in SAS/STAT (SAS Inst. Inc., 2001).

III – Results and discussion

The average warm carcass weight varied from 82.5 kg in the meaty group to 93.3 kg in KP (Table 1). The carcass weights of the KP and fatty groups were similar. Lean meat percentage was 47.8% in KP, and 51.3%, 57.9% and 64.0% in the fatty, normal and meaty groups, respectively. Thus, the differences among groups were largely caused by variations in back subcutaneous adipose tissue as well as muscle thickness. Lack of breeding is reflected in thicker backfat and thinner muscle compared to commercial pigs. Although pigs from meaty group were lighter than KP, they have 15 mm thicker muscle, and only one third the back subcutaneous adipose tissue thickness. The carcass weights of KP pigs were similar to those of the fatty group. However, the carcass traits describing body composition were in favour of the fatty group.

Table 1. Carcass traits in experimental groups of pigs

	KP (n=10)		Fatty (n=14)		Normal (n=15)		Meaty (n=14)	
	$\bar{x}$	±SD	$\bar{x}$	±SD	$\bar{x}$	±SD	$\bar{x}$	±SD
Carcass weight (kg)	93.3	±12.8	92.9	±11.1	88.3	±13.2	82.5	±9.4
Backfat thickness (mm)	33	±7	24	±5	16	±5	10	±2
Muscle thickness (mm)	61	±5	67	±7	72	±8	76	±9
Lean meat content (%)	47.8	±2.5	51.3	±2.4	57.9	±4.0	64.0	±3.2

KP – Krškopolje pigs.

Technological quality parameters (Table 2) showed no differences in pH value measured 24 hrs after slaughter between groups. Conductivity after 24 hrs in *m. longissimus dorsi* was smaller in KP (3.77 mS/cm) than by fatty (6.36 mS/cm) and meaty (6.40 mS/cm) commercial fatteners.

Result shows better technological quality of KP, because border threshold for normal quality is under 6 mS/cm (Blendl in sod., 1991). Krškopolje pigs had darker and more red meat in comparison with commercial fatteners (Table 2). The result was nearer to desirable value for pig meat 42 – 46 (PIC, 2003) in KP.

Intramuscular fat content in LD of Krškopolje and fatty pigs was around 2% (Table 2). The lowest content of IMF (1.4%) was observed in the meaty group. There were no differences in IMF between KP and the fatty and normal groups. Although KP had greater amount of adipose tissue pigs (Table 1), they do not accumulate IMF (Table 2), possible because barren environmental condition. Kuhn *et al*. (1997) compared a local German breed, the German Saddle Back with commercial Landrace pigs, finding that the former had twice the IMF content of the latter (2.87% and 1.33%). Result confirmed that indigenous breeds have a higher capacity for lipid deposition and are expected to have higher IMF content (Kuhn *et al*., 1997).

The nutritional quality of subcutaneous adipose tissue is important as backfat is widely used in manufactured meat products (Reichardt *et al*., 2003). In the current study, the proportion of SFA in the back subcutaneous adipose tissue did not vary between the groups (Table 3). The KP had the highest proportion of MUFA (50.8%) and a lower proportion of PUFA than the fatty group. It has been suggested that an adipose tissue of good nutritional quality should contain less than 15% PUFA (Houben and Krol, 1983). With 13% PUFA the subcutaneous adipose tissue of KP was fulfilled these suggested levels.

Table 2. Technological quality parameters of *m longissimus dorsi* and *m. semimembranosus* by KP and commercial pigs

Variable	KP (n=10)	Fatty (n=14)	Normal (n=15)	Meaty (n=14)	SEM	p - value
pH_{24} LD	5.49	5.65	5.57	5.54	0.07	0.3101
pH_{24} SM	5.57	5.87	5.77	5.74	0.09	0.0959
$COND_{24}$ LD (mS/cm)	3.77^{b}	6.36^{a}	5.80^{ab}	6.40^{a}	0.69	0.0222
$COND_{24}$ SM (mS/cm)	8.09	8.53	8.77	7.71	0.88	0.7608
L*	48.10^{b}	51.98^{a}	52.69^{a}	53.83^{a}	1.07	0.0016
a*	9.50^{a}	7.63^{b}	7.36^{b}	7.43^{b}	0.51	0.0085
b*	2.38	3.31	3.35	3.09	0.35	0.1562
c*	9.80	8.37	8.12	8.07	0.56	0.0944
h*	0.25^{b}	0.43^{a}	0.45^{a}	0.42^{a}	0.04	0.0010
IMF content (%)	1.96^{a}	1.94^{a}	1.70^{ab}	1.40^{b}	0.15	0.0462

KP – Krškopolje pigs; SEM – standard error of mean; COND – conductivity; LD – *m. longissimus dorsi*; SM – *m. semimembranosus*; IMF – intramuscular fat; superscripts within the same line are significantly different ($p < 0.05$)

Table 3. Nutritional quality parameters of subcutaneous adipose tissue by KP and commercial pigs

Variable	KP (n=10)	Fatty (n=14)	Normal (n=15)	Meaty (n=14)	SEM	p -value
Saturated fatty acids	36.13	38.48	39.24	37.63	0.97	*0.0781*
Monounsaturated fatty acids	50.81^{a}	43.35^{b}	44.70^{b}	43.96^{b}	0.85	<0.0001
Polyunsaturated fatty acids	12.72^{b}	17.85^{a}	16.05^{ab}	17.02^{ab}	1.40	0.0403
n-6 PUFA	11.64^{b}	16.56^{a}	14.85^{ab}	15.90^{ab}	1.29	0.0289
n-3 PUFA	0.97	1.19	1.08	0.97	0.12	0.5974
n-6 PUFA/n-3 PUFA	12.09^{b}	14.86^{ab}	14.77^{ab}	16.56^{a}	0.93	0.0087
PUFA/SFA	0.35	0.47	0.42	0.44	0.05	0.2522
Atherogenic index	0.44	0.47	0.48	0.48	0.02	0.2648

KP – Krškopolje pigs, SEM – standard error of mean; n-6 – omega 6; n-3 – omega 3; PUFA – polyunsaturated fatty acids; SFA – saturated fatty acids; superscript within the same line are significantly different ($p < 0.05$)

Differences among groups were also found in proportions of n-6 PUFA and in the n-6/n-3 PUFA ratio (Table 3). The KP contained 5% less n-6 PUFA than the fatty group. The n-6/n-3 PUFA ratio of the meaty group was significantly higher than the KP. Furthermore, the n-6/n-3 PUFA ratio of all the groups (Table 3) exceeded the nutritional recommendation of 4:1 (Enser *et al*., 2001). High n-6 PUFA compared to n-3 PUFA proportions in subcutaneous adipose tissue could be explained by the use of feed components rich in C18:2n-6, such us wheat and barley (Souci *et al*., 2000). Fatty acids n-3 PUFA are present in many feed ingredients but at lower levels than n-6 PUFA (Wood *et al*., 2008). There were no differences in P/S and AI indices among the groups (Table 3).

IV – Conclusions

The organically raised Krškopolje pigs had lower electrical conductivity, more dark and red colour of *m. longissimus dorsi* compared to commercial fatteners. Higher intramuscular fat content were observed in KP and fatty than meaty pigs.

Fatty acid composition of back subcutaneous adipose tissue of KP was nearer to the normal and meaty groups than fatty group. Krškopolje pigs had higher MUFA content than commercial fatteners, lower PUFA and n-6 PUFA than fatty group and lower n-6/n-3 index than meaty group.

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Evaluation of the fat content in a small-calibre Salami made with pork from Chato Murciano breed

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Abstract. Production of Chato Murciano breed, a Mediterranean rustic pig, requires the development of differentiated quality meat products with high commercial value. The aim of this study was to adjust the fat content of a small-calibre Salami manufactured with pork from Chato Murciano breed (castrated, 180 kg live weight and 18 months aged). Three types of Salami with different fat level estimated by near infrared probe (High 20%, Medium 18%; and Low 16% fresh mass) were processed according standard industrial practices, using, *Pediococcus pentosaceus* and *Staphylococcus xylosus* as starter culture, and *Penicilium crysogenum* as coverage moulds. The Salami was stuffed into pig casings and then were dried for 12 days (12-14 ° C and 90-75% RH). Several quality parameters were determined: proximate composition, drying-ripening rates (dehydration, acidification, fermentation, proteolysis, lipolysis and fat oxidation) and eating quality. The final content of fat was 27.7% for higher, 26.1% for medium and 24.2% for lower fat Salami. Lower fat favoured lactic fermentation and acidification (pH = 4.5), improved the reddening and intensified the proteolysis and the lipolysis. The intensity of aroma and taste of cured-fermented meat was similar in the fat range tested, since Salami showed small variations in firmness, but not in juiciness. Fat level did not affect the acceptance, so we recommend a final fat content of 24% for the Salami made with pork from Chato Murciano

Keywords: Salami – Chato Murciano – Fat.

Évaluation de la teneur en gras de la saucisse sèche du porc Chato Murciano

Résumé. *La production durable de porcs de races rustiques, comme le Chato Murciano, exige le développement de dérivés de viandes d'une qualité différenciée avec une haute valeur commerciale. L'objectif de l'étude a été d'établir le niveau en graisse idéal pour la saucisse sèche de porc de race Chato Murciano (castré, avec 180 kg de poids vif et 18 mois d'âge). Trois types de saucisses expérimentales furent élaborées avec de la viande maigre et 3 pourcentages de graisse différents estimés par proche infrarouge (élevé 20%, moyen 18% et faible 16% de masse fraîche). La saucisse fut élaborée avec un procédé industriel standard avec une culture initiatrice de* Pediococcus pentosaceus *et* Staphylococcus xylosus*, couverture de* Penicillium crysogenum*. Elle fut emboutie dans des tripes naturelles de porc et fut séchée pendant 12 jours (12-14ºC et 90-75% HR). La qualité est définie selon la composition, les indices de séchage-maturation (déshydratation, acidification, fermentation, protéolyse, lipolyse et oxydation des graisses) et l'analyse sensorielle. La teneur finale en graisse fut: 27,7% élevée, 26,1% moyenne et 24,2% faible. La faible teneur en graisse a favorisé la fermentation lactique et l'acidification correcte de la charcuterie (pH=4,5), a amélioré le rougissement et a intensifié la protéolyse et la lipolyse. L'intensité de l'arôme et de la saveur à viande séchée-fermentée fut similaire dans la frange de graisse testée, en observant de petites variations de fermeté et d'onctuosité, mais pas de jutosité. L'acceptation n'a pas été affectée par la teneur en graisse, c'est pourquoi nous recommandons 24% de graisse finale pour la saucisse de Chato Murciano.*

Mots-clés. *Saucisse – Chato Murciano – Graisse.*

I – Introduction

Chato Murciano breed is a Mediterranean rustic pig originated from Iberian trunk. In the early twentieth century this breed was of high socio-economic importance, being traditionally raised at home and in extensive systems with remnants of the orchard. Actually, Chato is being recovered by farmers of Murcia Region. Pigs were raised semi-extensively, fed a balanced diet based on special feeds and optionally local raw materials, and slaughtered around 18 months old and 180 kg live weight. Chato provides heavy carcasses and pork meat with high levels of haematic pigments, proteins, infiltration and oleic acid, being a suitable raw material for dry-cured products (Galián *et al*., 2008). The sustainable breeding of Chato Murciano would be given for the elaboration of differentiated quality meat products with high commercial value. Local companies from Murcia Region are actually developing pork sausages from Chato Murciano. An example is traditional small-calibre salami (less than 45mm diameter), ripened for 2-3 weeks, which is widely consumed in Spain. High fat infiltration of Chato pork offers the possibility to manufacture salami with no added back fat. This supposes certain technological risk, since fat strongly contributes to eating quality of salami, since fat intensifies the aroma, taste and juiciness. However, excessive fattiness can also limit the commercialization of salami, due mainly to nutritional aspects.

II – Materials and methods

Salamis were manufactured according industrial practices with pork from Chato Murciano breed. Three fat levels were tested (High 20%, Medium 18%; and Low 16% raw mass) (HF, MF and LF). Fat content was determined by near infrared (NIR) probe. The recipe (g kg^{-1}) of salami was: boned pork (880), water (44), sodium chloride (22), black and white pepper (10), dextrose, lactose and sucrose (20), dextrin (20), potassium nitrate (0.25) and sodium nitrite (0.25), sodium isoascorbate (0.5), sodium citrate (0.3) sodium glutamate (2.5) and Ponceau 4R red (0.2). Starter cultures of *Pediococcus pentosaceus*, *Staphylococcus xylosus* and *Penicilium Crysogenum* were used. The salami was stuffed into pig gut and then was dried for 12 days (12-14ºC and 90-75% RH). Several quality parameters were determined: proximate composition drying-ripening rates (dehydration, acidification, fermentation, proteolysis, lipolysis and fat oxidation) and eating quality. The effects of fat level on the quality of salami were determined by simple ANOVA.

III – Results

Table 1 shows the effects of fat level on the proximate composition and drying-ripening indices of salami. After drying, the fat content of salami reached percentages of 27.7% (high), 26.1% (medium) and 24.2% (low). Salami may contain up to 50% fat (Moretti *et al.*, 2004; Rubio *et al.,* 2008). Appropriated a_w value was reached for all three salamis, although HF had lowers moisture and a_w. Rates of ash and protein were similar in al three salamis, although the MF salami showed a slightly lower protein percentage. LF salami showed more pronounced acidification, reaching pH value of 4.5 at the end of ripening, while pH fell to 4.9 in HF and MF salamis. Reddening, proteolysis and lipolysis were more intense in LF.

Total viable and lactic acid bacteria counts were higher in LF than in MF and HF (Table 2). Low fat level also improved the lactic fermentation of Salami, due to the rate microbiological growth increase at higher a_w. Rapid acidification by LAB in LF was associated with lower counts of *Micrococaceae*.

Table 3 shows the effects of the fat level on the eating quality of Salami. Surprisingly, lean colour scoring was higher in HF and MF than in LF. The intensity of dry-cured colour may increase when Salami contains more lean meat and equal dose of curing agents. Major sensory attributes of Salami such as odour, flavour, juiciness and fattiness were not affected by the fat level, while minor differences in hardness were found as a function of fat level. Fat increased softness, although the overall acceptance was not affected. Thus, fat reduction can be made without loss of the eating quality of Salami. Acid flavour scoring did not agree with lactic acid content and pH found, getting the higher value in MF Salami.

Table 1. Effect of fat level on the proximate composition and drying-ripening of Salami prepared with different fat levels

	Fat level		
	High (M ± SD)	**Medium (M ± SD)**	**Low (M ± SD)**
Lipids NIR (g/100 g)	27.7±0.52 [a]	26.1±0.68 [b]	24.2±0.84 [c]
Moisture NIR (g/100 g)	34.3±0.33 [b]	38.4±0.87 [a]	37.9±1.43 [a]
Proteins NIR (g/100 g)	23.7±1.02 [a]	21.8±1.02 [b]	23.5±1.02 [a]
Ash (g/100 g)	5.4±0.33	5.3±0.33	5.3±0.33
a_w	0.89±0.01 [b]	0.91±0.01 [a]	0.90±0.01 [a]
pH	4.88±0.03 [a]	4.94±0.07 [a]	4.47±0.06 [b]
Lactic acid (g lactic 100 g^{-1})	0.67±0.02 [a]	0.51±0.03 [b]	0.67±0.07 [a]
Proteolysis (g NPN/100 g^{-1}TN)	11.08±1.42 [ab]	10.15±1.31 [b]	12.37±0.96 [a]
Fat acidity (mg KOH g^{-1})	4.43±0.36 [c]	6.87±1.11 [b]	14.36±1.94 [a]
L* Lightness (CIE units)	44.14±0.99 [b]	48.42±1.27 [a]	48.38±1.58 [a]
a* Redness (CIE units)	15.82±1.01 [b]	14.55±0.94 [b]	17.74±0.94 [a]
b* Yellowness (CIE units)	2.68±0.65 [b]	3.74±0.46 [a]	4.48±0.27 [a]
C* Chroma (CIE units)	16.06±1.09 [b]	15.02±0.98 [b]	18.29±0.95 [a]
º Hue (CIE units)	9.53±1.80 [b]	14.41±1.43 [a]	14.18±0.66 [a]

M: mean; SD: standard deviations; [a, b, c] Fat level effects (P≤0.05).

Table 2. Effects of fat level on the fermentative microflora (log fcu g^{-1}) of Salami prepared with different fat levels

	Fat level		
	High (M ± SD)	**Medium M ± SD)**	**Low (M ± SD)**
Total viable counts	8.83±0.14 [b]	8.72±0.08 [b]	9.20±0.09 [a]
Lactic acid bacteria	8.72±0.08 [b]	8.55±0.30 [b]	9.15±0.16 [a]
Micrococacceae	6.86±0.39 [ab]	7.33±0.41 [a]	6.45±0.23 [b]
Moulds and yeasts	5.16±0.15 [a]	5.32±0.33 [a]	4.45±0.22 [b]

M: mean; SD: standard deviations; [a, b, c] Fat effects (P≤0.05).

IV – Conclusions

Fat content of 24% (16% raw mass estimated by NIR) would be adequate to manufacture pork Salami from Chato Murciano breed, stuffed into natural gut and dried-ripened for 12 days. This fat reduction favoured the ripening properties of Salami, including lactic fermentation, without relevant loss of eating quality. Fat reduction could help with the commercialisation of Chato Murciano dry-cured sausages.

Table 3. Effects of fat level on the eating quality of Salami prepared with different fat levels

	Fat level		
	High (M ± SD)	Medium (M ± SD)	Low (M ± SD)
Lean colour	4.5±0.10 [a]	4.4±0.10 [a]	3.9±0.17 [b]
Odour	3.2±0.09 [b]	3.5±0.23 [a]	3.3±0.19 [ab]
Pepper odour	2.6±0.18 [ab]	2.4±0.21 [b]	2.8±0.16 [a]
Flavour	3.1±0.14 [b]	3.4±0.09 [a]	3.0±0.19 [b]
Pepper flavour	2.7±0.06	2.8±0.19	2.7±0.26
Acid flavour	3.0±0.19 [b]	3.9±0.13 [a]	3.1±0.15 [b]
Hardness	3.0±0.08 [b]	3.6±0.15 [a]	3.3±0.07 [a]
Juiciness	2.9±0.19	3.0±0.11	2.9±0.21
Fattiness	2.7±0.06	2.8±0.18	2.8±0.16
Acceptance	3.5±0.17	3.4±0.16	3.4±0.20

M: mean; SD: standard deviations; scoring scale: 1-5; [a, b, c] Fat effects (P≤0.05).

Acknowledgments

José Reverte SL (pig farmer) and Elaborados Cárnicos de Lorca SL (meat industry).

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Development of Chato Murciano sobrasada prepared using "Appellation of Murcia Origin" paprika

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Abstract. Sustainable production of Chato Murciano breed, a Mediterranean rustic pig, requires the development of differentiated quality meat products with high commercial value. The aim was to develop a sobrasada (a fatty dry-cured sausage) manufactured with Chato Murciano pork and AMOP. Sobrasada was prepared with pork from Chato Murciano (castrated and slaughtered at 180 kg live weight). Three types of paprika (Control: 6% standard; Mixed: 3% Standard + 3% AMOP; 6% AMOP) were tested. Sobrasada was stuffed into blind gut and was ripened for 30 days (14-18ºC and 75-80% RH). Quality parameters were determined: composition, aw, pH, colour L*a*b*, proteolysis, lipolysis, lipid oxidation, total viable counts, lactic acid bacteria, Micrococcaceae, yeasts, moulds, and eating quality. The use of AMOP hardly affected the technological characteristics of sobrasada, although proteolysis was more intense in samples containing 3% and 6% AMOP. The type of paprika mainly affected the eating quality. The use of AMOP intensified the typical aroma and taste of sobrasada, but provided less intense red-orange colour. It therefore seems advisable to mix AMOP with more pigmented paprika to achieve a good balance between bouquet and colour for sobrasada made with Chato Murciano pigmeat.

Keywords. Sobrasada – Chato Murciano – Paprika – Murcia.

Développement de la Sobrasada de Chato Murciano élaborée avec du paprika "Appellation d'Origine Contrôlée (AOC) de Murcie"

Résumé. *La production durable de porcs de races rustiques, comme le Chato Murciano, exige le développement de dérivés de viande d'une qualité différenciée avec une haute valeur commerciale. L'objectif fut de devélopper une Sobrasada de porc Chato Murciano élaborée avec du paprika "Appellation d'Origine Contrôlée (AOC) de Murcie". La Sobrasada fut élaborée avec de la viande et du lard de porc de race Chato Murciano (castré et abattu à 180 kg de poids vif). Trois types de paprika furent testés (contrôle: 6% standard, mélangé 3% standard + 3% AOC, et 6% AOC). La Sobrasada a été mise dans des boyaux de porc et a maturé pendant 30 jours (14-18ºC et 75-80% HR). Les paramètres de qualité déterminés furent: composition, aw, pH, couleur L*a*b*, protéolyse, lipolyse, oxydation des lipides, aérobies mésophiles totaux, bactéries acido-lactiques,* Micrococcaceae*, moisissures et levures et attributs sensoriels. L'utilisation de paprika AOC n'a presque pas influencé les caractéristiques technologiques de la Sobrasada, bien que la protéolyse ait été plus intense dans les échantillons avec 3% et 6% de paprika AOC. Le type de paprika affecte surtout la qualité sensorielle. L'utilisation de paprika AOC a intensifié l'odeur et la saveur caractéristique de la Sobrasada, alors qu'elle a donné une couleur rouge-orange moins intense. Donc, nous recommandons le mélange de paprika AOC avec du paprika plus pigmenté ou des oléorésines pour obtenir un bon équilibre entre arôme et couleur pour la Sobrasada fermentée de Chato Murciano.*

Mots-clés. *Sobrasada – Chato Murciano – Paprika – Murcie.*

I – Introduction

Chato Murciano breed is a Mediterranean rustic pig originated from Iberian trunk. Actually, Chato is being recovered by farmers of Murcia, SE Spain. Chato pigs were raised semi-extensively, fed a balanced diet based on special feeds and optionally local raw materials, and slaughtered around 18 months old and 180 kg live weight. The sustainable breeding of Chato

Murciano would be given for the elaboration of differentiated quality meat products with high commercial value. Local consumers and restaurants have begun to demand Chato pork once again and local companies are actually developing Chato sausages. Chato pig provides heavy carcasses with high fattening (Galián *et al.*, 2008), whose excess backfat must be transformed in meat products. Sobrasada, a fatty (40-50% fat) dry-cured sausage prepared with paprika, offers great opportunities in this sense. Paprika is also traditionally produced in the Region of Murcia, being obtained by mixing diferent varieties of *Capsicum annuum* (Rosselló *et al*., 1995). Depending of origin, paprika provides particular colouring and flavouring to meat products. Thus, two traditional ingredients from Murcia, Chato pork and paprika, can be used to produce a quality differentiated sausage. The aim was to develop Sobrasada manufactured with Chato Murciano pork and Appellation Murcia Origin Paprika (AMOP).

II – Materials and methods

Sobrasada was manufactured according industrial practices with pork from Chato Murciano breed. Three types of paprika (Control: 6% standard; Mixed: 3% Standard + 3% AMOP; AMOP: 6% AMOP) were tested. The recipe (g kg^{-1}) of sobrasada was: boned pork (365), backfat (325), paprika (60), odorous white wine (6), minor spices (oregano), salt and additives (60). Sobrasada was stuffed into blind gut and was ripened for 30 days (14-18ºC and 75-80% RH). Quality parameters were determined: proximate composition, a_w, pH, colour L*a*b*, proteolysis, lipolysis, lipid oxidation, total viable counts, lactic acid bacteria, *Micrococcaceae*, yeasts, moulds and eating quality. The effects of fat level on the quality of sobrasada were determined by simple ANOVA.

III – Results

Table 1 shows the proximate composition and drying-ripening indices of sobrasada manufactured with different paprika.

Table 1. Proximate composition and drying-ripening indices for Sobrasada from Chato Murciano prepared with different paprika (standard *vs* Murcia Origin)

	Paprika source		
	Control (M ± SD)	**Mix (M ± SD)**	**AMOP (M ± SD)**
Moisture (g 100 g^{-1})	29.5±1.54	29.6±0.92	29.3±1.36
Proteins (g 100 g^{-1})	12.9±1.55 ^{a}	8.65±1.06 ^{b}	10.5±2.65 ^{ab}
Lipids (g 100 g^{-1})	51.5±0.34 ^{b}	54.0±0.32 ^{a}	53.4±0.30 ^{a}
Collagen (g 100 g^{-1})	1.50±0.27	1.33±0.28	1.11±0.24
Ash (g 100 g^{-1})	5.44±0.33	5.41±0.31	5.47±0.32
a_w	0.90±0.01 ^{b}	0.89±0.00 ^{c}	0.92±0.00 ^{a}
pH	5.38±0.07	5.51±0.02	5.46±0.13
L* Lightness (CIE units)	48.4±0.72 ^{b}	48.6±0.35 ^{b}	50.3±0.52 ^{a}
a* Redness (CIE units)	35.7±1.33	37.1±0.51	35.9±0.61
b* Yellowness (CIE units)	29.2±2.76	30.5±0.85	31.0±1.57
C* Chroma (CIE units)	49.6±6.01	48.0±0.86	47.4±1.45
º Hue (CIE units)	39.2±1.81	39.4±0.60	40.8±1.04
Proteolysis (g NPN 100 g^{-1} TN)	15.9±2.30 ^{b}	22.9±1.76 ^{a}	23.0±6.28 ^{a}
Fat acidity (mg KOH g^{-1})	19.0±0.88	20.3±2.21	18.2±0.72
TBARS (mg MDA kg^{-1})	0.19±0.02	0.18±0.02	0.18±0.04

Paprika sources: 6% standard (Control) : 3% Standard + 3% AMOP (Mix); 6% AMOP (AMOP). M: mean; SD: standard deviations; $^{a, b, c}$ Paprika effects (P≤0.05).

After drying, slight differences in proteins and lipids were found for different types of sobrasada. Agreeing this, a_w value was slight higher in AMOP than in Mix and the Control sobrasadas. Surprisingly (reddening war higher in standard paprika), no chromatic differences were found between the Control, Mix and AMOP sobrasada, although AMOP showed the highest L* value. The interaction between paprika and other ingredients, especially fat, may explain these results for colour. On the other hand, sobrasada presented higher proteolysis than the Control and Mix sobrasadas, while no effect of paprika source on fat acidity and lipid oxidation were found. The low counts found for the main fermentative groups (Table 2) indicated that no relevant fermentation take place during the ripening stage.

Table 2. Main fermentative groups (log fcu g^{-1}) for sobrasada from Chato Murciano prepared with different paprika (Standard *vs* Murcia Origin)

	Paprika source		
	Control (M ± SD)	**Mix (M ± SD)**	**AMOP (M ± SD)**
Total viable counts	5.78±0.05	5.83±0.04	5.85±0.13
Micrococacceae	3.90±0.31^{ab}	3.95±0.06^{a}	3.44±0.41^{b}
Lactic acid bacteria	5.55±0.26	5.43±0.03	5.46±0.02
Moulds and yeasts	5.73±0.12	5.84±0.21	5.72±0.16

Paprika sources: 6% standard (Control) : 3% Standard + 3% AMOP (Mix); 6% AMOP (AMOP).
M: mean; SD: standard deviations; $^{a, b, c}$ Paprika effects (P≤0.05).

Table 3 shows the sensory scores of sobrasada manufactured with different paprika. Sobrasada was characterized by a typical red-orange colour. In opposite to CIEL*a*b* values, the trained panellists detected colour differences between AMOP-Mix and the Control samples. The intensity of colour was lower scored in AMOP sobrasada than in the Control sobrasada, while Mix sobrasada obtained intermediate scoring. However, the aroma and taste scores were higher in AMOP-Mix than in the Control sobrasada. The use of AMOP at different concentration (3% and 6%) clearly intensified the sobrasada aroma and taste. These results suggest that AMO paprika provided better flavouring and certain discolouration to sobrasada. The physical-chemical, microbiological and sensory data obtained were coherent with those obtained from preliminary studies on Stabilization by chilling of sobrasada from Chato Murciano pigmeat manufactured without preservatives (Martínez *et al.*, 2009).

IV – Conclusions

The use of AMOP hardly affected the technological characteristics of sobrasada, although proteolysis was more intense in samples containing 3% and 6% AMOP. The type of paprika mainly affected the eating quality. The use of AMO paprika intensified the characteristic aroma and taste of sobrasada, but provided less intense red-orange colour. It therefore seems advisable to mix AMOP with more pigmented paprika to achieve a good balance between bouquet and colour for sobrasada prepared with Chato Murciano pigmeat.

Table 3. Sensory scoring for sobrasada from Chato Murciano prepared with different paprika (Standard *vs* Murcia Origin)

	Paprika source		
	Control M ± SD	**Mix M ± SD**	**AMOP M ± SD**
Colour	3.5±0.6 [a]	3.2±0.4 [ab]	2.9±0.5 [b]
Colour homogeneity	2.9±0.6	2.9±0.7	2.8±0.7
Aroma	3.0±0.6 [b]	3.5±0.5 [a]	3.4±0.4 [a]
Paprika aroma	2.8±0.6	2.9±0.7	2.8±0.7
Acid aroma	1.8±0.6	1.7±0.6	1.8±0.6
Taste	2.9±0.6 [b]	3.4±0.5 [a]	3.5±0.5 [a]
Paprika taste	3.0±0.6	3.3±0.6	3.2±0.6
Acid taste	1.7±0.5	1.8±0.5	1.9±0.5
Bitter aftertaste	2.2±0.8	2.3±0.8	2.1±0.5
Fattiness	3.4±0.8	3.6±0.4	3.7±0.4
Creaminess	3.6±0.7	3.8±0.4	3.8±0.5
Fibrous residue	2.3±0.6	2.3±0.6	2.4±0.6

Paprika sources: 6% standard (Control) : 3% Standard + 3% AMOP (Mix); 6% AMOP (AMOP).
M: mean; SD: standard deviations; [a, b, c] Paprika effects (P≤0.05).

Acknowledgements

José Reverte S.L. (pig farmer) and Elaborados Cárnicos de Lorca SL (meat industry).

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The weight loss in the production of non-fermented salami "*capocollo*", "*fiocco*" and dry cured ham from "*casertana*" pig ancient autochthonous genetic type (AAGT). Further contribution

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Abstract. The study was carried out to monitor the weight loss of three unfermented local products (LP), "capocollo Sannita", "fiocco Sannita" and dry cured ham, obtained from males (castrated not less than 40 days before slaughter) and entire females belonging to the "Casertana" pig AAGT, reared at the experimental farm of ConSDABI Sub NFP.I.-FAO. The results, valid within the observation field, showed that for all three products the weight loss during the seasoning, in the same conditions, was statistically greater (P<0.001) in castrated male than that obtained from the entire female and, in particular: (i) the capocollo Sannita [54 (♂♂) and 62 ♀♀] has decreased by 25% *vs* 20% at 1 month, 39% *vs* 33% at 3 months and 45% *vs* 42% at 6 months; (ii) the fiocco Sannita [51 (♂♂) and 41♀♀] has decreased by 22% *vs* 17% at 1 month and 43% *vs* 39% at 12 months; (iii) the dry cured ham [34 (♂♂) and 42 ♀♀] has decreased by 9% *vs* 7% at 1 month, 18% *vs* 14% at 3 months reaching the highest value at 24 months (33% *vs* 27%).

Keywords. Ancient autochthonous genetic type (AAGT) – Capocollo Sannita – Fiocco Sannita – Dry cured ham – Local Products (LP).

Diminution de poids lors de la production de "capocollo", "fiocco" et "prosciutto" issus du type génétique autochtone ancien (TGAA) "Casertana". Des contributions supplémentaires

Resumé. *L'étude a été menée pour surveiller la diminution de poids pendant la maturation de trois produits locaux (PL) non fermentés : "capocollo Sannita", "fiocco Sannita" et "prosciutto Sannita", obtenus à partir de mâles (castrés au moins 40 jours avant l'abattage) et femelles non castrées du TGAA "Casertana" (CT) élevés chez le ConSDABI SUB NFP.I.- FAO. Les résultats, valables dans le champ d'observation, ont montré que pour les trois produits la perte de poids pendant la maturation, en conditions égales, s'est avérée plus élevée pour le mâle castré par rapport à la femelle non castrée et, en particulier: (i) le "capocollo Sannita" [54 (♂♂) et 62 ♀♀] a diminué de 25%* vs *20% à 1 mois, de 39%* vs *33% à 3 mois et de 45% vs 42% à 6 mois ; (ii) le 'fiocco Sannita' [51 (♂♂) et 41♀♀] a diminué en moyenne de 22% vs 17% après 1 mois, et de 43% vs 39 après 12 mois de maturation ; (iii) le "prosciutto Sannita" [34 (♂♂) et 42 ♀♀] a diminué en moyenne de 9% vs 7% après 1 mois, de 18% vs 14% après 3 mois et de 33% vs 27% après 24 mois.*

Mots-clés. *Type génétique autochtone ancien (TGAA) – Capocollo Sannita – Fiocco Sannita – Prosciuto Sannita – Produits locaux.*

I – Introduction

Casertana (CT) pig, also called "pelatella", for the absence of bristles, or "napoletana" for its place of origin, is one of the best Italian autochthonous pig population so that Höesch (first half of 10th century) defined it as "pig Italian pride". During the centuries this population underwent alternate events. It contributed, in the past (half of 19th century) to the development of the Yorkshire and Berkshire English breeds, while at the end of 80's years its population size decreased so that it became a genetic type at risk of extinction. Currently, CT pig is reared in Campania, Lazio, Molise and Umbria regions, with a growing population size. Not fermented products, economically more important, obtained from this AAGT are without any doubt "ham" and "fiocco". The aim of this research was to furnish a further contribution to the knowledge of the influence of gender factor on the weight loss during the seasoning of Local product (LP) obtained from meat of CT pig.

II – Materials and methods

The study involved unfermented local products (LP): "capocollo Sannita", "fiocco Sannita" and dry cured ham obtained from males (castrated less than 40 days before slaughter) and entire females of the Casertana pig AAGT. The animals, reared in multiple boxes at experimental Farm of ConSDABI Sub NFP.I. - FAO, were fed with commercial feed. Net live weights of pigs at the slaughter were 168.8 kg and 163.6 kg respectively for castrated male and entire female. Scheme 1 reports the number of the products analyzed and the period in which the weight variation (weight loss) was registered for each 'LP'.

In particular: (i) the capocollo Sannita was made using neck (starting from atlas-occipital articulation to the 5th thoracic vertebra) after trimming; its seasoning takes about six months; (ii) fiocco Sannita, the noble part of the ham, consisting of three muscles (*Semimembranosus*, *Semitendinosus* and *Biceps femoris*) has a peculiar 'pear' shape (typical of "culatello of Zibello"); its seasoning takes about 12 months; (iii) the dry cured ham was obtained from the leg of pig properly prepared until to give it the typical rounded shape; its seasoning takes about 24 months.

The seasoning was realized in proper places controlled for temperature and humidity. Each product was weighted at the start of the process, at the end of drying time, and then periodically (weekly or monthly) until the end of seasoning time (Table 1). Seasoning weight loss was calculated as: [(initial product weight– product weight during process)/ initial product weight]*100.

No preservatives (nitrite and nitrate) were used.

Table 1. Sequence of monitoring for each product

Product	(♂♂)	♀♀	Seasoning d									
			0	6	15	30	90	180	270	360	540	720
Capocollo	54	62	X	X	X	X	X	X				
Fiocco	51	41	X			X	X	X		X		
Ham	34	42	X			X	X	X	X	X	X	X

The preliminary statistical analyses showed a significant influence of initial weight of product on the trend of weight loss, so the data were processed using the following model of covariance analysis in which initial weight of the product was the covariate and 'gender' was considered as fixed factor (SAS, 1997):

$y_{ijk} = \mu + b_1x_1 + gender_i + e_{ijk}$; where:
μ = constant common to all the observations (general mean);
b_1 = regression coefficients of dependent variable on the weight loss of product (x_1);

$gender_i$ = fixed effect of i^{th} gender (i = 1, 2);
e_{ijK} = random error and/or unknown effects.

The Student's t test was applied to compare the estimated means.

III – Results and discussions

The results, valid within the observation field, showed the significant influence of gender to determinate the variation of weight of all three considered products. Tables 2, 3 and 4 evidenced that the products obtained from meat of entire females had a significantly lower weight loss, in comparison with those obtained from meat of castrated males (*P*<0.001). In particular, for the capocollo Sannita this difference reached the higher value (5.6%) after 90 days of seasoning, as well as fiocco Sannita (6.3%), while for dry cured ham the highest difference was observed at 360 days (6.7%).

Table 2. Percentage weight loss of "capocollo Sannita" during seasoning period

Seasoning d	Gender		Δ = [(♂♂) - ♀♀]
	(♂♂)	♀♀	
3	5.32	4.55	0.77***
15	16.15	13.41	2.74***
30	24.67	20.17	4.50***
90	38.84	33.26	5.58***
180	45.38	41.59	3.79***

****P<0.001.*

Table 3. Percentage weight loss of "Fiocco Sannita" during seasoning period

Seasoning d	Gender		Δ = [(♂♂) - ♀♀]
	(♂♂)	♀♀	
3	8.75	8.18	0.57**
14	14.47	12.78	1.69***
30	21.58	17.34	4.24***
90	33.80	27.52	6.28***
180	40.30	34.18	6.12***
360	43.04	38.66	4.38***

***P<0.01 ***P<0.001.*

Table 4. Percentage weight loss of dry cured ham during seasoning period

Seasoning d	Gender		Δ = [(♂♂) - ♀♀]
	(♂♂)	♀♀	
30	9.04	6.86	2.18***
90	18.50	13.78	4.72***
180	24.49	18.30	6.19***
270	26.62	20.17	6.45***
360	28.59	21.85	6.74***
540	31.30	24.66	6.64***
720	33.19	27.16	6.03***

****P<0.001.*

At the and of seasoning time, the average weight loss of dry cured ham was 30.18%, lower than Istrian ham (46.31%) (Karolyi *et al*., 2005) or Bayonne ham (from 35% to 39%) (Monin *et al*., 1997) and not more different from Parma ham (about 27%) (Nanni Costa *et al*., 1999).

IV – Conclusions

The results, valid within the observation field, highlighted that for the three local products considered, the castrated male had a significantly greater percentage weight loss than entire female. This trend confirms the results obtained by Castellano *et al*. (2006). These authors report that the difference could be attributed to a different texture of fat tissue or to a higher content in intramuscular fat of entire females (not published data). The relation with qualitative data (rheology and colour) taken on both muscular portion and on covering fat of the dry cured ham and fiocco Sannita will may provide useful indications in order to deep factors that influence these differences.

Acknowledgements

The authors wish to acknowledge the Mipaaf for financial support and they wish to also thank dressed pork factory "Di Maria", Cese Alte – Circello (BN).

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The weight loss in the production of dry cured sausages "salsiccia" and "soppressata" from *"Casertana"* pig ancient autochthonous genetic type (AAGT). Further contribution

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Abstract. The study involved 605 "soppressata Sannita" and 121 "salsiccia Sannita" obtained processing meat of Casertana (CT) AAGT pigs [68 castrated males (castrated not less than 40 days before slaughter) and 53 entire females] reared at experimental farm of ConSDABI Sub N.F.P.I.-FAO in order to evaluate the effect of gender on weight loss of the ripened products. The results, valid within the observation field, showed a significant effect of gender on weight loss of both products. In particular: (i) the soppressata Sannita obtained from castrated males had a percentage weight loss greater in comparison with the meat of entire females; at the end of seasoning time (40 days) it lost (P<0.01) about the 51% of initial weight (+4% in comparison with that of entire female); (ii) the salsiccia Sannita that at 15 days had a percentage weight loss of 41% if obtained from meat of castrated males and of 34% if obtained from meat of entire female (P<0.05), after 30 days had a weight loss of 52% and 47%, respectively (P<0.05).

Keywords. Casertana pig – Ancient autochthonous genetic type (AAGT) – Salsiccia Sannita – Soppressata Sannita.

Diminution de poids lors de la production de "salsiccia" et de "soppressata" issus du porc de type génétique autochtone ancien (TGAA) "Casertana". Des contributions supplémentaires

Résumé. *L'étude a concerné 605 "soppressate sannite" (saucisson) et 121 "salsicce sannite" (saucisse) préparés avec de la viande de chaque porc [68 mâles (castrés au moins 40 jours avant l'abattage) et 53 femelles non castrées] du TGAA "Casertana", élevés chez le ConSDABI SUB NFP.I.-FAO afin d'évaluer l'effet du sexe sur la diminution de poids des deux produits fermentés. Les résultats, valables dans le champ d'observation, ont montré une significative influence du facteur sexe sur le taux de perte de poids pour les deux produits. En particulier : (i) la soppressata Sannita préparée avec la viande de mâle castré a un pourcentage de diminution constamment plus grand comparé à celui obtenu en utilisant la chair de femelle non castrée ; à la fin du séchage (40 jours) au total on perd (P <0,01) 51% du poids (+4%) par rapport à la femelle non castrée ; (ii) la salsiccia Sannita après 15 jours montre une perte de poids de 41% lorsqu'elle est produite avec de la viande de mâle castré et de 34% lorsqu'elle est produite avec de la viande de femelle non castrée (P <0,05), après 30 jours on perd 52% et 47% respectivement (P <0,05).*

Mots-clés. *Casertana – Type génétique autochtone ancien (TGAA) – Salsiccia Sannita – Soppressata Sannita.*

I – Introduction

"Casertana" (CT) in the past was reared in particular in Campania and in neighbouring regions and was appreciated, besides for its good productive performances, also for its tendency to accumulate fat. This particular characteristic was one of the reasons for which this AAGT was replaced with pigs of foreign breeds, selected to make more lean meat. The recent attention to the conservation of the genetic resources and the high demand for local products had allowed to a renewed productive utilization of this AAGT. The valorisation of local products is a desirable and feasible target for cultural, scientific, economic and social reasons. The local products, different from industrial products for ancient origin and for specific organoleptic characteristics, allow to safeguard productive techniques and traditions, to improve the conditions of rural populations, in particular in marginal areas and also to preserve the AAGTs from the eventual extinction. This work is a part of a project aimed at enhancing the use and production of CT pig AAGT; in particular, in this note, we report the results of the monitoring of ripening process evolution of two products, soppressata Sannita and salsiccia Sannita, which enhance the ancient Italian tradition of meat and especially sausage consumer, monitoring the weight loss for 30 and 40 days respectively.

II – Materials and methods

The study involved 726 individual products obtained from 68 males (castrated at least 40 days before slaughter) and 53 entire females, slaughtered at average live weight of 169 kg and 161 kg respectively. All animals were reared in multiple boxes at experimental farm of ConSDABI Sub NFP.I.- FAO and were fed with commercial feed. The products were made at a salami factory. For the preparation of these LPs the meat was minced and mixed; pH, temperature and humidity at different seasoning times were registered. The seasoning was realized in proper places controlled for temperature and humidity during the 24 hours. Each product was weighed at the end of the drying time and then periodically (daily or weekly) until the end of seasoning time. The dough for the preparation of soppressata Sannita consisted of separable meat obtained from the partial trimming of ham and long back with the addition of lumbar subcutaneous fat derived from trimming of ham, cut manually with knife in an amount equal to about 2- 3% of the total weight of the used meat; for the preparation of salsiccia Sannita, separable meat from jowl, diaphragm, capocollo trimming, partial trimming of ham, shoulder, track, trimming of belly, ribs filet and throat was used.

The length of fresh soppressata Sannita must be about 18 – 20 cm, with a minimum regular circumference of 18 cm when it was fresh.

The length of fresh salsiccia Sannita must be about 50 – 60 cm and it has a typical form of horseshoe bat.

The seasoning included a drying phase (7 days) at controlled temperature (14-18°C) and relative humidity (60-70%) and a second seasoning period (45 and 30 days, respectively for soppressata Sannita and salsiccia Sannita) at lower temperature (10-14°C) and higher relative humidity (70-85%).

At the end of seasoning all two products, after regular brushing and washing with lukewarm water to remove the moulds, were vacuum-packed or preserved under pork fat. The data were processed using the following model of covariance analysis, with initial weight of product and net live weight as covariate for soppressata Sannita and weight of cooled carcass and age at slaughter as covariate for salsiccia Sannita (SAS, 1997):

$Y_{ijK} = \mu + b_1x_1 + b_2x_2 + gender_i + e_{ijk}$, where:

μ = constant common to all the observations (general mean);

b_1, = regression coefficients of the dependent variable from net live weight of pig or weight of ‘cooled carcass’ (x_1);

b_2, = regression coefficients of the dependent variable from initial weight of product or ‘age at slaughter’ (x_2);

$gender_i$ = fixed effect of i^{th} gender (i = 1,2);

e_{ijk} = random error and/or unknown effects.

The significance of differences between the estimated means was tested using Student's t test.

III – Results and discussion

The weight loss during the seasoning time was significantly higher in all two products obtained from castrated male in comparison with that obtained from entire female; in particular (Tables 1 and 2).

(i) for soppressata at the end of seasoning (44 days) the difference reached 4% (P<0.001), starting from 1.9% at 5 days (P<0.01);

(ii) 'salsiccia Sannita' obtained from castrated males registered about 41% of weight loss at 15 days of seasoning (P<0.05) and 52% at the end of seasoning (P<0.05), values higher than that obtained from entire females (about 34% and 47% respectively).

Table 1. Soppressata Sannita. Percentage weight variation in relation to the seasoning time and gender

Seasoning d	Sex		Δ = [(♂♂) - ♀♀]
	(♂♂)	♀♀	
5	20.4	18.4	1.9**
10	28.0	25.4	2.6***
15	34.3	31.2	3.1***
20	38.2	34.5	3.6***
30	47.6	43.7	3.9***
44	51.4	47.4	4.0***

***P<0.01; ***P<0.001*

On the average from meat of each castrated male it was obtained 27.5 kg of salsiccia Sannita while from meat of each female it was obtained 23.3 kg. This difference is due to the different live weight of each gender (169,2 *vs* 160.9 kg).

Table 2. Salsiccia Sannita. Percentage weight variation in relation to the seasoning time and gender

Seasoning d	'Sex'		Δ = [(♂♂) - ♀♀]
	(♂♂)	♀♀	
15	40.8	33.6	7.2*
30	51.8	47.2	4.6*

**P<0.05.*

Some researchers (Nold *et al*., 1999; Maiorano *et al.,* 2007) showed that meat obtained from sow has a higher water holding capacity respect to meat of male, and this can be one of the reason that had determined the different weight loss observed by us.

IV – Conclusions

The results, valid within the observation field, highlighted that, for the two considered local products, the castrated male had a greater percentage weight loss than entire female. This trend may confirm previous results (Barone *et al*., 2006). This difference could be due to the different adipose tissue's texture (for a probable different amount of saturated fatty acids), as well as to the higher intramuscular fat content and a best water holding capacity of the meat obtained from entire females. This result in association with qualitative data of product (rheology and color traits) may be suggest a diversification of the products also in the selling price on the bases of gender of the pig that provide raw material.

Acknowledgements

The authors wish to acknowledge the Mipaaf for financial support and they wish to also thank dressed pork factory 'Di Maria' Cese Alte – Circello (BN).

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Selection of staphylococci strains isolated from a Portuguese traditional fermented/dry sausage for potential use as starter cultures

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Abstract. In order to evaluate its potential use as starter cultures, 104 isolates of *Staphylococcus* spp. were obtained from sausages and different sampling environment points in workshops A and B, at southern region of Portugal. PCR amplification was carried out to confirm genus and species allocation. From workshop A, 47 isolates were staphylococci, the majority belonging to the specie *S. equorum*, while from workshop B, of the 57 isolates identified as staphylococci, most were *S. xilosus*. The genetic profiles of isolates were further compared using PCR fingerprinting analysis, guiding to the selection of 43 representative strains subsequently characterized for their nitrate reductase, lipolytic and proteolytic activities. Among these strains, 30% revealed proteolytic ability while 42% had lipolytic activity. 65% of the strains reduced nitrate. Subsequently, 10 strains (representatives of different fingerprinting groups) were evaluated for their ability to grow at different temperatures, pH and NaCl concentrations. *S. xylosus* and *S. equorum* showed to growth under all the studied conditions being more effective at 15°C, 20°C and pH 5.5 at 20°C, and NaCl concentrations of 10% and 15%; therefore can be guaranteed their application in technological processes with varying temperatures. In workshop A, strains P05-58 *S. carnosus* and P05-74 *S. equorum* and, from workshop B, P06-01 and P06-26 *S. xylosus* presented the most interesting features.

Keywords. *Staphylococcus* coagulase negative – Fermented meat products – Starter culture – Proteolytic activity – Lipolytic activity – Nitrate reductase activity.

Sélection de souches de staphylocoques isolées à partir de produits carnés traditionnels portugais fermentés pour leur utilisation potentielle comme culture starter

Résumé. *Afin d'évaluer leur potentielle utilisation comme culture starter, 104 isolats de* Staphylococcus *ont été obtenus à partir de "chouriços" et de différents points de l'environnement des unités A et B, au sud du Portugal. La confirmation génétique du genre et de l'espèce a été réalisée par PCR en identifiant 47 isolats de staphylocoques dans l'unité A, dont la plupart ont été* S. equorum*, tandis que dans l'unité B les 57 isolats étaient en majorité* S. xilosus*. Par PCR fingerprinting on a sélectionné 43 souches, qui ont été caractérisées concernant les activités nitrate réductase, lipolytique et protéolytique. De ces souches, 30% ont montré une activité protéolytique, 42% avaient une activité lipolytique, et 65% avaient la capacité de réduire les nitrates. Par la suite, 10 souches de différents groupes de fingerprinting ont été évaluées pour leur capacité de croissance à différentes températures, pH et % de NaCl.* S. xylosus *et* S. equorum *ont montré une croissance dans les conditions étudiées, étant plus efficaces à 15°C et 20°C, et dans un pH de 5,5 à 20°C, et dans des conditions de NaCl de 10% et 15%; ainsi on peut garantir leur application dans des procédés technologiques avec des températures variables. Les souches P05-58* S. carnosus *et P05-74* S. equorum *de l'unité A, autant que les* S. xylosus *P06-01 et P06-26, de l'unité B, ont démontré être les plus intéressantes.*

Mots-clés. *Staphyloccocus coagulase négative – Produits carnés fermentés – Culture starter – Activité protéolytique – Activité lipolytique – Activité nitrate réductase.*

I – Introduction

In Portugal there is a wide variety of traditional fermented/dry meat products, produced in small scale units being characterized by having in their manufacture a spontaneous fermentation by means of the so-called "house flora". Such products suffer a slight acidification and sensory characteristics are highly appreciated by consumers. Chorizo is a sausage made with pork, slightly fermented, smoked and dried, with a low pH and a decreased water activity, its production is characterized by a close relationship with the fermentative flora naturally present in the industrial environment. This type of flora is introduced in the meat during slaughter and increased in its concentration during manufacture of the fermented product. Each processing unit has a specific environmental flora, composed of useful microorganisms for the fermentation and the development of sensory characteristics in traditional sausages. The role of *Staphylococcus* as technological culture is well defined and the selection of the most interesting strains is a challenge to improve the hygienic and sensory characteristics of traditional sausages.

This study aimed to characterize the technological flora of *Staphylococcus* spp. isolated from dry fermented sausages and the traditional manufacture environments, in order to assess their potential use as starter cultures.

II – Material and methods

1. Origin of *Staphylococcus* isolates

The experimental work was carried out using 104 staphylococci isolated from the manufacturing environment, samples of traditional fermented and smoked sausage (chorizo) and secondary ingredients (garlic, pimento paste or bell pepper paste and natural casing) at two traditional meat industries (Plants A and B) located in Alentejo. The isolates were previously identified by biochemical miniaturized tests APIStaph (Biomerieux, France) by Fraqueza *et al.* (2006). At each manufacturing plant, samples were taken from the work surfaces (table, knife, grinder, mixer or stuffing machines) and from the fermented/smoked sausages in three different stages of manufacture (product after filling, product with half time smoking period and the final product) and secondary ingredients.

2. Extraction of DNA

The isolates were grown in tryptose salt agar to obtain fresh culture by 24 h incubation at 37°C, and the extraction of genomic DNA was performed using the QIAamp DNA Mini Kit 02/2003" from Quiagen (Germany).

3. Identification

The isolates were identified as belonging to the species *Staphylococcus aureus, Staphylococcus epidermidis*, *Staphylococcus xylosus* and *Staphylococcus saprophyticus* using a multiplex PCR method described by Morote-Bizot *et al.*, (2004). *Staphylococcus carnosus* and *Staphylococcus simulans* were identified by PCR performed according to the methodology described by Blaiotta *et al.*, (2004) while the identification of *Staphylococcus equorum* was based on the protocol described by Blaiotta, *et al.*, (2004).

4. Genetic characterization and strain selection

The selection of isolates to assess for technological interest was based on their genetic profiles using the PCR fingerprinting methodology according to Tenreiro (2007). Representative strains were selected by comparison of the fingerprinting profiles obtained.

5. Phenotypic characterization of technological functions

The nitrate reductase activity was determined as described by Miralles *et al* (1996), while proteolytic activity was performed by sowing among PCA with gelatin, peptone and milk powder with subsequent flooding of the plate with a solution of mercuric chloride after incubation. To evaluate the lipolytic activity we used the Spirit Blue Agar medium (France).

Selected representatives (n = 10) groups with different genetic profiles were characterized for their ability to grow in different conditions of temperature, pH values and sodium chloride concentrations in order to evaluate their potential use as starter cultures. We investigated 3 incubation temperatures (10°C, 15ºC and 20°C), two pH values (5.0 and 5.5), the temperatures of 15 and 20ºC and two different concentrations of sodium chloride (10 and 15%).

6. Statistical analysis

Descriptive analysis of the results and their graphical presentation was obtained using Microsoft Office Excel 2007. The fingerprinting profiles obtained for each isolate were introduced in the Bionumerics software (version 4.61 [Applied Maths, Kortrijk, Belgium]) and dendrograms were constructed using the Pearson correlation coefficient and clustering method based on the unweighted average distance (UPGMA – unweighted pair group method with arithmetic average).

III – Results and conclusions

In Plant A, forty-seven Staphylococcus isolates were identified as members of the species *S. equorum* while in Plant B, fifty-seven isolates belong to the species *S. xylosus.* The genetic profiles of the isolates were compared by PCR fingerprinting (Figs 1 and 2) and led to the selection of 43 staphylococci which were subsequently characterized for their nitrate reductase, lipolytic and proteolytic activities.

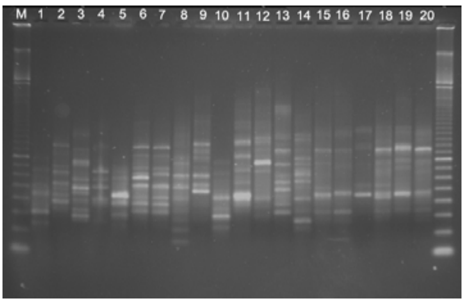

Fig. 1. PCR fingerprinting profiles of *Staphylococcus* strains.

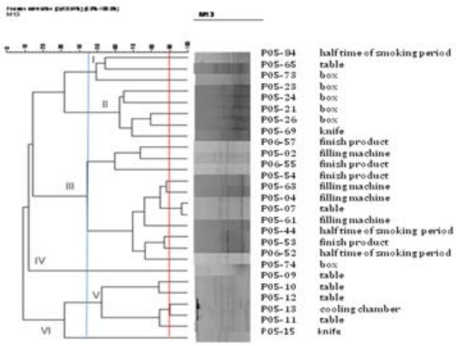

Fig. 2. Dendrogram analysis of the genetic profile of strains of *Staphylococcus equorum.*

By analyzing Fig. 2 we can see that the genetic profiles clustered the isolates in three main groups, with similarities below 45%. Strains P05-04 and P05-07 harbor profiles with a genetic similarity above 90%, being considered identical or highly related.

It was found that 30% of the strains showed proteolytic activity, while 42% showed lipolytic activity. As for the nitrate reductase activity, it was observed that about 65% of the strains reduced nitrate.

Strains of *S. xylosus* and *S. equorum* (n = 10) grew in all the conditions studied, but their development was more effective at 15°C and 20°C, pH 5.5 at 20°C and 10% and 15% concentration of chloride sodium, thus suggesting features that can guaranteed their application in technological processes with varying temperatures (Table 1)

Table 1 . Evaluation of proteolytic, lipolytic and nitrate reductase activity in strains of *Staphylococcus xylosus* and *Staphylococcus equorum*

	***S. xylosus* (n=10)**		***S. equorum* (n=8)**	
	Positive	**Negative**	**Positive**	**Negative**
Proteolytic Act. (n=18)	3	7	3	5
Lipolytic Act (n=18)	10	-	1	7
Nitrate reductase Act. (n=18)				
30ºC/7 h (n=18)	5	5	-	8
20ºC/24 h (n=18)	6	4	7	1
15ºC/72h (n=18)	2	8	7	1

IV –Conclusions

Strains of *S. carnosus* P05-58 and *S. equorum* P05-74 from Plant A and strains of *S. xylosus* P06-01 and P06-26 from Plant B, demonstrated by their characteristics to be the most interesting from the technological point of view. These results suggest their potential usage as starter cultures.

Acknowledgements

This work was financially supported by EU programe QLK1-CT2002-0240 and CIISA.

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Quality of the sirloin "presa" of the Iberian pork in two types of package

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Abstract. Availability of meat from Iberian pork, which is allowed to graze free range Dehesas (acorn forests) giving pigs fat its unique sweet *flavour*, is limited as its production is seasonable. Freezing may be an option to prolong commercialization periods and further consume of this meat, which represents high economic value. Aim of this study is to compare the quality of frozen meat, "presa" packed under vacuum in PA/PE material and frozen meat packed under vacuum in PA/PE material and stored in carton boxes, taking refrigerated meat as a reference. For this study, 24 samples were freeze using different storage conditions, namely 12 samples were packed individually under vacuum, and 12 samples were packed individually under vacuum and laid up in cardboards. Reference samples were prepared from same batch and stored at refrigerating conditions.After a conservation period of 5 month at -18°C, samples were thawed and its physicochemical properties (pH, color, moisture, water retention capacity), texture (Texture Profile Analysis, penetration) and sensorial analysis (performed by a trained panel), were assessed with the aim of evaluating possible damages caused by freezing. The results show evidences on differences between refrigerated packaging systems. Frozen meat delivered a lower pH and was darker then refrigerated meat which was confirmed by a decrease on L*. Cut resistance was as well lower for frozen meat. After thermal treatment, moisture content decreases followed by a decrease on meat hardness and adhesiveness. Sensory analysis points out a decrease on *flavour* persistence, overall acceptance, and aftertaste, with significant differences being verified on meat packed only in PA/PE material.This results evidence that packing Iberian pork meat under vacuum and cardboards is the packaging alternative which delivers meat with closest characteristics to the ones observed on refrigerate meat.

Keywords. Iberian pig – "Presa" – Meat – Freezing – Packaging.

La qualité de la longe ("presa") du porc Ibérique surgelée sous deux formes d'emballage

Résumé. *La disponibilité de la viande de porc Ibérique élevé en chênaie, est fonction du rythme saisonnier de sa nourriture (glands). La congélation permet de prolonger sa commercialisation et par conséquent la consommation de cette viande, d'une valeur économique supérieure. Cette étude vise à comparer la qualité de la longe ("presa") du porc Ibérique surgelée en pochette, ainsi que celle de la même viande surgelée en pochette et placée en boîte, par rapport à la viande réfrigérée, et à évaluer le processus de congélation. Nous avons utilisé 24 échantillons surgelés, 12 échantillons emballés individuellement sous vide et 12 échantillons emballés sous vide mais placés en boîtes et 12 échantillons réfrigérés, du même lot, qui ont servi comme référence. Après 5 mois de conservation à une température de -18ºC, les échantillons ont été décongelés et leurs propriétés physiques et chimiques (pH, couleur, humidité, capacité de rétention de l'eau), leur texturation (analyse du profil de la texture, test de pénétration simple) et leurs propriétés sensorielles (panel de dégustateurs chevronnés), ont été évaluées, afin d'apprécier les altérations provoquées éventuellement par la congélation. Les résultats démontrent qu'il existe des différences entre les formes d'emballage, notamment la diminution du pH, la diminution de la valeur L* et la diminution de la résistance à la coupe. Après un traitement thermique, on a constaté une diminution de l'humidité, de la dureté et de l'adhésivité. L'évaluation sensorielle démontre que l'acceptation globale, la persistance de la saveur propre à la viande et le goût résiduel ont diminué, et présentent une différence significative, ceci uniquement dans les échantillons surgelés en pochette. On arrive ainsi à la conclusion que la congélation en pochette avec empaquetage en boîte est la modalité qui permet le mieux à la longe (« presa ») surgelée, de se rapprocher de la longe réfrigérée.*

Mots-clés. *Porc Ibérique – Longe ("Presa") – Congélation – Surgelé – Emballage.*

I – Introduction

The production of the Iberian pork may be considered seasonal as there are some offering peaks. During the periods of higher distinct production, freezing will be a way of storing the product and so there is a guarantee of adjusting to the market. In industry the portions of a greater economic value were commercialized fresh. When there were no freezing processes, the meat portions were almost used in the manufacture of smoked sausages. The aim of this work is testing the freezing method of the ("presa" / sirloin or portion) ventral, serriform, toraxic and cervical muscles of the Iberian pork used in the enterprise "Damicarnes". As we are dealing with meat of high economic value, it is supposed to evaluate the differences between the refrigerated and the frozen product in two distinct forms of package which create differences in the freezing speed of the product. Among all the characteristics of the quality of the meat, texture is extremely important for the consumer (Lawrie,1998).

The texture of the meat corresponds to sensations of succulence and tenderness which are evaluated by instrumental techniques or by a panel of tasters. The greatest difficulty in evaluating the meat texture is, no doubt, establishing a relationship between the results obtained by the two methods, and above all, because the meat has to suffer a thermal treatment before being consumed, which increases the impact in the meat characteristics (Genot, 2003). Freezing is a preservation system and the technology used tries to limit the damages aiming at causing the slightest number of alterations (Varnan and Sutherland, 1998). Package may confer distinct degrees of protection to the product. The development of flexible pellicles and vacuum package led to the evolution of the meat distribution sector. Meat frozen after vacuum package, keeps its natural quality during a more prolonged period (Price and Schweigert, 1994). The main causes for the deterioration of fat are hydrolysis and oxidation. It is of the utmost importance to freeze as fast as possible after production, under the correct conditions and reduce to the minimum the temperature fluctuations during storage and transportation (Varnan and Sutherland, 1998).

Freezing is the best way to preserve meat at a long term but it must be well done. The maintenance of the characteristics of the product during preservation depends on the temperature of the storage and the type of meat (Prandal *et al*., 1994) The loss of CRA and the lack of capacity the fibres have to reabsorb the water when thawing and then high ionic force causes denature of the muscle proteins (Lawrie, 1998). The quantity of exsudation varies according to the initial characteristics of the product, the speed of freezing and thawing, the temperature and the period of storage, however, this quantity is lower when the meat is vacuum stored with an impermeable material to oxygen and the flavour of rancidity also decreases. According to Genot (2003), the freezing usually causes a slight increase in the tenderness of the meat, and the force of the muscle cutting which suffered freezing is much lower than in the fresh meat. Pork is slightly more tender and juicy, when it is frozen and it is also more easily chewed. An excessive loss of exsudation is a determining factor in the quality of the meat and unpleasant to the consumer, restricting the acceptability of the product (Varnan and Sutherland, 1998).

II – Material and methods

The sample used in this study is the sirloin of the Iberian pork. The sirloin is made up of the serriforme, ventral toraxic and cervical near the thigh (Mayoral *et al.*, 2003). It is considered a sample, a portion of meat which corresponds to the sirloin and weighs approximately 700 g with the dimensions 20 x 10 x 5 cm, which suffered a process of freezing in a current of forced air and picked at hazard from the right and left side of males and females. Thirty six examples of the sirloin of the Iberian pork were collected which came from the same lot of meat from animals which underwent identical conditions of handling and slaughter and were vacuum stored at +2ºC. The plan of sampling followed in this study is the following: 6 samples were used in laboratory analyses and the remaining 6 in a sensorial analysis. The sirloin was packed unit by unit, in vacuum with a poliamid polyethylene pellicle of 125 μm thickness in a thermoformative machine and frozen according to Table 1. After freezing it was moved to the preservation chamber of frozen products.

Table 1. Plan of samples

No. of samples	Refrigerated samples	Samples frozen in a vacuum pouch under a shelf (0ºC to -7ºC) 4h30; 5 months at -18ºC	Samples frozen in a vacuum pouch and packaged in carboard boxes under a shelf; (0ºC to -7ºC) 12h; 5 months at -18ºC
6+6	CR		
6+6		CBL	
6+6			CCX

pH was determined according to the rule NP 33441 / 1990. The colour was determined by a colorimeter previously calibrated by a plate with a white reference no. 19733057, channel 0 (L*= 97.10; a*= + 0.07; b*= +1.83). Result expressed in: L*,a*,b*; Relationship a/b; Chromaticity C= $(a^2 + b^2)0.5$;Tone Hº = arctangent b/a* 360º / (2*3.14)(Minolta, 1991). The percentage of humidity (moisture) was determined according to the procedure of the rule NP 1614/2002. The capacity of water retention was carried out according to the pistometric method (Grau and Hamm, 1953). The rheologyc characterization was carried out in a texturometer. The samples were submitted to tests of a simple cutting. Slices of the muscle were cut, 2 cm length in the vertical and each one in parallelepipeds of 1.5 cm. They were grilled in a convector oven at 150ºC, during 20 minutes, the grilled meat was kept in a stove at about 55ºC until the experiment. The test was carried out with a chopping knife and the speed of the test was 1 mm s^{-1} the distance of 10 mm. The test was carried out with a thermal treatment and without a thermal treatment (in the direction of the muscular beam and against it). The characteristics determined were the resistance to the cutting and the work of the strength of the cutting.

The sensorial evaluation of the Iberian pork was tested by a panel of tasters selected and trained by the Agrarian High School in Beja , according to the NP ISO 8586-1 (2001). Tasting occurred according to NP 4258 (1993). To carry out the experiments, slices of meat were cut with 2 cm width and each slice in parallelepipeds of 1.5 cm and they were grilled in a convector oven.

III – Results and discussion

The average values of pH in the refrigerated meat are superior and with significant differences compared to the values found in the frozen meat inside a pouch and inside a pouch and a box which also show significant differences (Table 2) The average values of CRA don't show significant differences in the different ways of package of the meat frozen in a pouch and a box compared to the refrigerated meat. However, the values showed clearly a fall. Bustabad (1999) refers that vacuum package and the package in cardboards is effective and contributes to reducing the loss of water.

The values of moisture in the meat before being submitted to a thermal treatment don't show any significant differences, however, they don't include the loss of water suffered by the exsudation of the thawed meat (which was about 3%). Farouk *et al*. (2003) showed that the losses of water when thawing were identical to the freezing speeds either high or low, when evaluated after 6 months preservation and increase all along the preservation period. The values obtained in the moisture of the meat after thermal treatment, show significant differences between the refrigerated meat, frozen in a pouch and frozen in a pouch and a box. Under the same conditions under the thermal treatment, the refrigerated meat, managed to keep the water in a more outstanding way than the meat which was frozen. Lawrie (1998) refers that a pH of 5.9 causes cooking losses superior to the losses when the pH is 6.0, that's to say that the reduction of pH caused an increase of water losses during cooking in a continued way.

Table 2. Averages, pattern deviations and results of the analysis of the variation of the physic chemistry parameters of the sirloin of the Iberian pork refrigerated (CR) frozen in a pouch (CBL) and frozen in a pouch and a box (CCX)

	CR	CBL	CCX
pH	6.17[a] (0.15)	5.93[b] (0.06)	5.85[c] (0.11)
CRA %	32.83[a] (2.12)	29.67[a] (7.14)	30.00[a] (3.84)
Meat moisture without thermal treatment	70.85[a] (1.43)	70.77[a] (0.86)	69.45[a] (2.87)
Meat moisture after thermal treatment	57.67[a] (0.36)	54.68[b] (0.46)	50.56[c] (0.18)

The Table 3 shows the values obtained in the colour of the muscle in the lateral face and in the medial face. It is verified that the meat lost its brightness, got dark after freezing and thawing packed in a pouch or in a pouch and box when compared to the values obtained in the refrigerated meat.

Table 3. Averages, patterns deviations and results of the analysis of the variation of the colour of the muscle in the outside lateral face and medial face, L*,a*,b*, (CIE) of the sirloin of the Iberian pork refrigerated (CR) frozen in a pouch (CBL) and frozen in a pouch and box (CCX)

	CR	CBL	CCX
L*	38.96[a] (2.60)	30.98[b] (3.46)	32.27[b] (2.32)
a*	17.72[a] (0.97)	15.67[b] (2.12)	18.76[a] (1.12)
b*	4.36[a] (1.04)	4.21[a] (1.84)	4.44[a] (1.19)
a/b	4.28[a] (1.07)	4.31[a] (1.52)	4.48[a] (1.08)
$C=(a^2 + b^2)^{0.5}$	18.27[a] (1.11)	16.28[b] (2.24)	19.30[a] (1.28)
H° = arctang b/a	0.24[a] (0.05)	0.25[a] (0.09)	0.23[a] (0.05)
L*	42.47[a] (4.16)	32.81[b] (3.33)	33.72[b] (3.59)
a*	17.98[a] (2.48)	18.01[a] (1.75)	18.06[a] (1.78)
b*	5.15[a] (1.76)	4.55[a] (1.17)	4.85[a] (1.36)
a/b	4.20[a] (2.75)	4.34[a] (1.74)	4.02[a] (1.22)
$C=(a^2 + b^2)^{0.5}$	18.75[a] (2.689)	18.62[a] (1.60)	18.74[a] (1.90)
H° = arctang b/a	0.28[a] (0.08)	0.25[a] (0.07)	0.26[a] (0.07)

In the study carried out by Estevéz *et al*. (2003) , the values of L* fell down just like it happened in this study, which can be explained by the water losses after thawing and hemoglobin concentration which reduces the value of L *. According to Price and Schweigert (1994) if the temperature of freezing is superior to -57ºC it is usually produced a dark colour because metamioglobin is formed. Because of the fall of the pH and CRA , the colour gets dark owing to the passage of mioglobin and metamioglobin (Farraia da Graça,1987) and the effect is greater with values of pH superior to 5.8 (Varnam and Sutherland 1998).

In all the tests of a simple cut in meat without thermal treatment it was observed that the cut resistance and the work of the cut strength showed lower values with significant differences, in the frozen meat inside a pouch or inside a pouch and box, when compared to refrigerated meat (Table 4)

The cellular destruction owing to the formation of intercellular ice crystals, led to the reduction of the cut force in the thawed meat (Lagerstedt *et al*., 2008). The denatured proteins are particularly sensitive to the attack of proteolitic enzymes, causing the reduction of hardness (Lawrie ,1998).

Table 4. Averages, pattern deviations and results of the analysis of variance for the parameters of the test of a simple cut in the sirloin of the Iberian pork refrigerated (CR), frozen inside a pouch (CBL), and frozen inside a pouch and a box (CCX), without thermal treatment (direct or against the muscular beam), and with thermal treatment

	Without thermal treatment for the muscular beam			Without thermal treatment against the muscular beam			With thermal treatment		
	CR	CBL	CCX	CR	CBL	CCX	CR	CBL	CCX
Cut resistance (N)	51.45^{a}	29.27^{b}	28.09^{b}	50.73^{a}	36.48^{b}	38.04^{b}	45.61^{a}	38.83^{ab}	31.92^{b}
	(35.79)	(7.58)	(8.72)	(17.63)	(10.45)	(11.23)	(27.06)	(5.56)	(9.79)
Work cut strength (NS)	107.94^{a}	72.00^{b}	66.13^{b}	140.02^{a}	97.29^{b}	104.82^{b}	121.64^{a}	133.20^{a}	100.95^{a}
	(46.11)	(23.03)	(24.22)	(46.14)	(32.25)	(40.70)	(75.44)	(20.96)	(39.09)

The average values in the same line differ when affected with different letters to ≤,0,05.

The values obtained in the cut for the muscular beam show a greater difference between the frozen meat and the refrigerated meat than in the values against the muscular beam, and the joints are more easily destroyed in freezing. According to Farag *et al*. (2009) another factor that influences the texture of the meat is the direction of the muscle fibres. After the thermal treatment it was checked that the cut resistance decreased especially in the meat which suffered freezing compared to refrigerated meat. The panel tasters detected significant differences in the residual taste of the frozen samples in the pouch when compared to the refrigerated ones and with those frozen in a pouch and box (Table 5). For an intermediate freezing speed, the ice is formed outside and inside the cell together with a variable temperature during preservation and leads to damages in the tissues. The CRA of the frozen meat in a pouch is inferior to the one frozen in a pouch and box, which becomes evident in a greater quantity of lost liquids during the thermal treatment , being drawn some precursors of the residual taste that represents the detection of remaining substances (Chamorro and Losada, 2002). In the global appreciation the panel of tasters considered that the samples frozen in a pouch show significant differences from the refrigerated ones but were not significant among the frozen samples.

Table 5. Average, pattern deviations and results of the analysis of variance for the sensorial parameters in the sirloin of the Iberian pork refrigerated (CR), frozen in a pouch (CBL), and frozen in a pouch and box (CCX)

	CR	CBL	CCX
Tenderness	5.66^{a}(0.62)	4.79^{a}(0.21)	5.61^{a}(0.15)
Succulence	5.38^{a}(0.24)	3.93^{a}(0.61)	4.29^{a}(1.11)
Flavour of rancidity	0.44^{a}(0.16)	0.79^{a}(0.11)	0.82^{a}(0.16)
Characteristic *flavour*	6.15^{a}(0.28)	5.68^{a}(0.16)	5.79^{a}(0.60)
Persistence	5.72^{a}(0.12)	4.93^{b}(0.10)	5.04^{b}(0.15)
Residual taste	5.13^{a}(0.12)	4.36^{b}(0.10)	4.93^{a}(0.20)
Global appreciation	6.64^{a}(0.43)	4.50^{b}(0.20)	5.57^{ab}(0.51)

The average values in the same line differ when affected with different letters to ≤,0,05.

IV – Conclusions

Freezing in a pouch and box obtained a better result than freezing in a pouch. In the existing conditions it isn't the speed of freezing that interferes in the quality of the frozen meat; this means that the method used at present in the enterprises is the most advisable, however it is important to stabilize the preservation temperatures to minimize the undergone changes. The frozen meat after being thawed and compared to refrigerated meat, shows an inferior aspect, a decrease in pH caused by the development of lactic acid bacteria, a darkness caused by the formation of metamioglobin, a decrease of CRA as well as moisture, hardness , adherence and cut resistance, caused by proteic denature.The tasters classified negatively the residual taste of the frozen sample in a pouch in comparison to the refrigerated one and to the frozen one in a pouch and box. In the global appreciation, tasters preferred refrigerated meat which they consider not showing significant differences with the frozen meat in a pouch and box, only manifesting significant differences in the frozen meat in a pouch. Weight loss during freezing and the storage of frozen meat oxidative and colour changes in meat from three lines of free-range reared Iberian pigs slaughtered at 90 kg live weight and from industrial pig during refrigerated storage.

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Characterization of surface mycoflora in Nebrodi hams

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Abstract. Within the framework of a three-year project, the screening of spontaneous fungal species grown on hams produced with "Nebrodi black pig" meat and seasoned in different environments of the Nebrodi area has been carried out, in order to characterize mycological population of such typical products and to keep under the presence of unexpected changes due to fungal contamination. In most of the aitchbones examined, the prevailing fungal species isolated resulted to be xerotolerant or xerophilic, due to their peculiar ability of adaptation to surface a_w and to thermohygrometric conditions applied in ripening rooms. With regard to the *Eurotium* strains isolated, *Eurotium herbariorum, Eurotium rubrum* and *Eurotium cristatum* have been detected. With regard to the *Penicillium* strains isolated, *Penicillium nalgiovense*, *Penicillium chrysogenum, Penicillium griseofulvum, Penicillium olsonii,* and *Penicillium aurantiogriseum* have been detected. In any of the aitchbones examined, undesirable *Penicillium* species such as *P. commune* or *Penicillium solitum* (which are considered responsible for the production of the so-called "phenol defect" in hams) and *P. nordicum* (which proved to be one of the greatest Ochratoxin A producer in hams and in protein-based foods) have not been detected.

Keywords. Phenol defect – Hams – Moulds – *Penicillium solitum – Penicillium nordicum.*

Caractérisation de la flore fongique de surface dans les jambons de Nebrodi

Résumé. *On a étudié la mycoflore sur les jambons produits avec la viande de porcins de races autochtones appartenant aux types génétiques Nebrodi et séchés dans les environnements de la région de Nebrodi, afin de caractériser la population fongique sur ces produits locaux et de surveiller la présence de changements anormaux dus à la contamination des moisissures. Dans la plupart des os du bassin («quasi») examinés, les espèces fongiques prédominantes ont été xérotolérantes ou xérophiles, grâce à leur capacité caractéristique de s'adapter à l'*a_w *superficielle et aux paramètres hygrothermiques utilisés dans les environnements de vieillissement. En ce qui concerne les espèces* Eurotium *isolées, on a constaté la présence de* Eurotium herbariorum*,* Eurotium rubrum *et* Eurotium cristatum*. En ce qui concerne les espèces de* Penicillium *isolées, ont été trouvées* Penicillium nalgiovense*,* Penicillium chrysogenum*,* Penicillium griseofulvum*,* Penicillium olsonii*, et* Penicillium aurantiogriseum*. Dans aucun des os du bassin examinés, n'ont été trouvées d'espèces indésirables de* Penicillium*, comme* P. commune *ou* P. solitum *(qui sont responsables du dénommé "défaut de l'acide phénique" dans les jambons) et* P. nordicum *(qui s'est avéré être l'un des plus grands producteurs d'ochratoxine A dans les jambons et les produits à base de protéines).*

Mots-clés. *Défaut de l'acide phénique – Jambons - Flore fongique – «Quasi» -* Penicillium solitum - Penicillium nordicum.

I – Introduction

Among factors influencing air-borne contamination of foods, microbial population in environmental air (Heldman, 1974) play a great role. In general, fungal spores proved to represent the prevailing part (56%) of the air-borne microflora (*Aspergillus*, *Penicillium*, *Rhizopus*, *Cladosporium*, *Fusarium*), while *Bacillus* spores, Gram-positive and Gram-negative bacteria represent the remaining one. Nevertheless, in industrial environments where foods such as meat derivatives are produced, fungal population can reach 70% of the total microbial population (Singh *et al.*, 1986).

In aged meats, moulding is directly connected with physico-chemical parameters recorded in productive environments (Baldini *et al*., 2000). In ripened meats, maturing techniques applied usually allow for the rapid and distinctive colonization by a great number of mycetes, which are considered fundamental to impart both desirable appearance and good organoleptic characteristics to these meats. On the contrary, in dry-cured meats maturing techniques are more and more pointed to obtain products where no or little surface mycoflora has grown: only autochthonous moulds indicating that ripening process is shaping up good and proving to compete and then prevail over undesired species should be tolerated. In general, in hams unexpected changes can occur and even persist in the final product in case thermo-hygrometric parameters reach high values during the resting and dehydration process is not carried out correctly.

A first example of such changes is represented by the so-called "phenol defect", a detrimental effect due to a fungal colonization of the aitchbone area by *Penicillium commune* (Spotti *et al*., 1988) or by *Penicillium solitum* (Spotti, personal communication) during seasoning in case salt content on ham surface results lower than 17.5% (saturated salt solution) during salting and Relative Humidity (RH) values result higher than 85%. In fact, this area represent the wettest part of the ham and it could be more subjected to spoilage by *Penicillium* species, while the muscle portion, where dehydration occurs in a faster way, is usually more subjected to spoilage by *Eurotium* species, which tend to prevail because of their xerophily (Baldini and Spotti, 1995). To avoid "phenic acid defect" it should be taken into account that: (i) a satured salt solution on the cut surface allows salt penetration in the aitchbone area during salting; (ii) fast dehydration is essential within the first 15 days of resting; (iii) the control of thermo-hygrometrical parameters such as RH is fundamental to avoid fungal growth; (iv) an increasing in dehydration could be applied in case fungal spoilage occurs during resting and pre-ripening.

A further example of the above-mentioned changes is represented by the presence in seasoned hams of ochratoxin A (OTA), a strong, nephrotoxic, secondary metabolite that can be produced in meat products by some fungal species such as *Aspergillus ochraceus* and *Penicillium nordicum* and that can persist in the finished product. The moulds responsible for OTA production can develop on the surface of aged products and start producing OTA in case some variations in thermo-hygrometric parameter occur, so their presence must be always kept under control by means of periodical laboratory tests.

Within the framework of the above-mentioned project, the aim of this work was to screen spontaneous fungal species grown on aitchbones from hams produced with "Nebrodi black pig" meat and seasoned in different environments of the Nebrodi area, in order to characterize mycetical population of such typical products and to keep under the presence of the above-mentioned unexpected changes due to fungal contamination.

II – Materials and methods

The screening of the mycoflora grown on aitchbones from hams seasoned in traditional environments of the Nebrodi area has been carried out by using sterile swabs, since they allow to scratch out a significant amount of conidia from any of the aitchbones assessed. After collecting the conidial mass, each swab has been plated on Malt Extract Agar (MEA) and on Dichloran 18% Glycerol Agar (DG18). Petri Dishes have been incubated at 25°C for seven days, in order to allow sporification of the fungal species isolated. Fungal identifications have been carried on selective media, according to the methods proposed by Pitt (Pitt and Hocking, 2009) and by Samson (Samson *et al*., 2004):

- Malt Extract Agar (MEAB), 25% Glicerol Nitrate agar (G25N), Creatine Sucrose Neutral Agar (CSN), CY20S, Yeast Extract Sucrose Agar (YES) incubated at 25°C for seven days;
- Czapek Yeast extract Agar (CYA) incubated at 5°, 25° and 37°C for seven days.

III – Results and discussion

The screening of the mycoflora grown on aitchbones from products seasoned in traditional environments of the Nebrodi area has been carried out on hams at different stages of their long-term ripening, since thermo-hygrometric parameters recorded in ripening plants proved to be greatly influenced by environmental outdoor conditions. In most of the aitchbones examined, yeasts have been isolated. In fact, they usually can colonise surface layers of cured meats for most of the seasoning time, both contributing to the development of the final typical aroma and avoiding oxidative processes on the ham surface (Martin *et al*., 2006).

In all the aitchbones examined, (Table 1) the prevailing fungal species isolated resulted to be xerotolerant or xerophilic (capable of growing at water activity (a_w) values lower than 0.85). In particular, the presence of *Eurotium* species and the growth of more xerotolerant fungal species belonging to *Penicillium* can be both due to their peculiar ability of adaptation to surface a_w and to thermo-hygrometric conditions of these plants where RH values range from 85 to 92% and where temperatures range from 10° and 20°C.

Table 1. Prevalence of fungal species isolated on aitchbones at the end of the seasoning from Nebrodi hams, at different stages of their long-term ripening

Species isolated	Frequency of contaminated aitchbones (%)		
	Year 2005	**Year 2006**	**Year 2007**
Eurotium herbariorum	85.7	63.6	38.5
Penicillium gladioli	71.4	18.0	46.0
Penicillium griseofulvum	71.4	27.0	7.6
Penicillium olsonii	57.0	9.0	23.0
Penicillium chrysogenum	0	9.0	7.6
Penicillium nalgiovense	0	9.0	7.6
Penicillium aurantiogriseum	0	27.0	23.0
Eurotium rubrum	0	9.0	30.7
Eurotium cristatum	0	18.0	7.6
Aspergillus candidus	0	9.0	0
Aspergillus versicolor	0	9.0	7.6
Penicillium glabrum	0	0	7.6
Aspergillus sydowii	0	9.0	0
Hyphopichia burtonii	0	9.0	0
Yeasts	85.7	72.7	100.0

With regard to the *Eurotium* strains isolated, *Eurotium herbariorum, Eurotium rubrum* and *Eurotium cristatum* have been detected. Their presence can be explained by the fact that: they're markedly xerophiles (so they're able to grow within a wide range of temperature and a_w values); they tend to form fast-growing colonies that usually prevail over *Penicillium* ones; they reproduce themselves in both a vegetative (via darkish-coloured conidia) and a sexual (via yellowish-coloured cleistotecia containing ascospores) way. Such species, as well as *Aspergillus* ones, can frequently occur on ripened and dry-cured meat, as the surface a_w is markedly lower on these kind of products.

With regard to the *Penicillium* strains isolated, *Penicillium nalgiovense*, *Penicillium chrysogenum, Penicillium griseofulvum, Penicillium olsonii,* and *Penicillium aurantiogriseum* have been detected. In literature, such species are frequently mentioned as those occurring in seasoning environments, so their presence has been well tolerated.

Among *Penicillium* species*, Penicillium gladioli* has been also detected. As it was isolated yet within the first part of the project on Nebrodi salami and it was focused on because of its properties as autochthonous "starter" culture, its presence has been well tolerated too.

In addition to this, it must be underlined that some undesirable *Penicillium* species such as *P. commune* or *Penicillium solitum* (which are considered responsible for the production of the so-called "phenic acid defect" in hams) and *P. nordicum* (which proved to be one of the greatest Ochratoxin A producer in hams and in protein-based foods) have never been detected on the examined aitchbones. This means that variations in thermo-hygrometric parameter haven't occurred and that seasoning have been carried out correctly in hams from Nebrodi area.

With regard to other environment-contaminating moulds, contamination by *Cladosporium, Aureobasidium* and *Mucor* species has been only rarely detected. The growth of these moulds should be avoided both on hams and salami, as they don't allow homogeneous drying of the product in the first steps of the process and they can form darkish spots on casings of salami or on surface of hams.

Ultimately, *Hyphopichia burtonii* has been detected just once. This species is attributed to Filamentous Fungi and indicated as "yeast-like mould" because of its morphological and reproductive characteristics which enable it to grow widely on the surface of solid media with a_w values ranging from 0.85 to 0.90. Its presence is usually tolerated as it proved not to produce toxic metabolites, to compete with undesired species and to partially inhibit OTA production (Spotti *et al*., 2009). For the above-mentioned reasons, its use as possible "competitor" of any undesired *Penicillium* species has been at present taking into account.

Aknowledgements

The authors wish to acknowledge the financial support received from the region Sicily. This research has been carried out within the framework of the project "Valorizzazione del sistema suinicolo dei Nebrodi. Difesa della biodiversità zootecnica come fattore di sviluppo del territorio. SP1: Caratterizzazione dei prodotti di salumeria macinati e interi ottenuti da carni di suino Nero dei Nebrodi: individuazione dei fattori di tipicità e sperimentazione tecniche di ottimizzazione delle produzioni". Regione Sicilia – IX Servizio Regionale allo Sviluppo.

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Comparison of pork quality from pure and crossbred Iberian pig

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Abstract. The Iberian (IB) Pig Breed is the most important Mediterranean swine type, both in population size and economic importance. Most of IB pork is consumed as cured products. However, the consumption of fresh meat has recently increased. Due to the increasing demand of fresh meat, in 2007, a new National Quality Standard (NQS) was published in Spain to regulate the production and marketing of products derived from IB pig carcass, including for first time fresh meat. This Quality Standard included two genetic product types in Iberian pork production: Iberian purebreed pork and Iberian x Duroc crossbreeding pork. In fact, Iberian x Duroc (50%) is the most common Iberian crossbreeding pig found in the meat market included in NQS. We have studied the main meat quality parameters of tenderloin (*psoas major* muscle) and *serratus ventralis* muscle, which are the most expensive meat cuts for fresh consumption, from those two genetic pig groups. Meat from IB pig showed different characteristics of that from crossbred pigs. However, more differences were observed in tenderloin than in *serratus ventralis* muscle. Tenderloin from crossbred pigs had lower water holding capacity, intramuscular fat and PUFA contents, and higher SFA content than tenderloin from Iberian purebred pigs. *Serratus ventralis* muscle from crossbred pigs had lower myoglobin content than *serratus ventralis* from IB purebred pigs, but no important differences were observed in other meat quality parameters.

Keywords. Meat quality – National Quality Standard – Iberian Pig.

Comparaison de la qualité de la viande entre la race Ibérique pure et croisée avec Duroc

Résumé. *Le porc Ibérique (IB) est la race porcine de type méditerranéen la plus importante, autant pour ses effectifs que pour son importance économique. La majorité des produits ibériques sont consommés sous forme de produits secs. Toutefois, la consommation de viande fraîche a augmenté récemment. En raison de la demande accrue de viande pour la consommation, en 2007 a été publiée une nouvelle norme de qualité pour réguler la production et le commerce des produits ibériques, incluant pour la première fois la viande fraîche. Dans cette norme de qualité existent deux types génétiques : produits de porcs purs Ibériques et produits de porcs croisés entre Ibérique et Duroc ; le produit croisé à 50% est le croisement le plus fréquent concernant la commercialisation, inclus dans la norme de qualité. Nous avons étudié les principales caractéristiques de qualité de la viande de l'aloyau (Psoas major) et du muscle Serratus ventralis, qui sont les viandes les plus chères, pour ces deux types génétiques. La viande de pur Ibérique a des caractéristiques différentes de celle des porcs croisés. Cependant, il y a plus de différences dans l'aloyau que dans le muscle Serratus ventralis. L'aloyau des porcs croisés a une moindre capacité de rétention d'eau CRA, une moindre infiltration de graisse intramusculaire, ainsi que des teneurs plus faibles en graisse et acides gras polyinsaturés PUFA, et supérieures en acides gras saturés SFA par rapport à la viande de pur Ibérique. Le muscle Serratus ventralis de porcs croisés contient moins de myoglobine que la viande provenant d'Ibérique pur, mais aucune différence significative n'a été trouvée pour les autres paramètres de qualité de la viande.*

Mots-clés. *Qualité de la viande – Normes de qualité – Porc ibérique.*

I – Introduction

The Iberian pig breed is the most important Mediterranean swine type, both in population size and economic importance. Traditionally, most of Iberian pork is destined to become dry-cured products. However, the consumption of several fresh meat cuts has recently increased, reaching high prices. Due to the increasing demand of fresh meat cuts, in 2007, a new National Quality Standard (NQS) for Iberian products was published in Spain to regulate the production and marketing of products derived from Iberian pig carcass, including for first time fresh meat, instead of dry-cured products only (dry-cured ham, dry-cured shoulder and dry-cured loin) (RD 1469/2007, of November 2). From the point of view of racial origin, that NQS included two genetic product types into Iberian pork production: Iberian Purebred pork and Iberian x Duroc crossbreeding pork. In fact, Iberian x Duroc (50 %) is the most common Iberian pork production found in the meat market included into the NQS. However, into NQS in adapting to Council Directive 88/661/EEC of December 19, the label "Iberian Purebred Pork" is restricted only to the Iberian products from livestock registered in the Studbook, which is a fraction of the total breed. As a result, products from Iberian pigs not registered in the Studbook are sold along with products from Iberian x Duroc crossbreeding, which are labelled as "Iberian" into NQS, without a commercial differentiation between these in the market. This creates a permanent discussion about the appropriateness of the explicit commercial differentiation of products from Iberian x Duroc crossbreeding.

We have studied the main meat quality parameters of tenderloin (*psoas major* muscle) and *serratus ventralis* muscle from those two genetic groups (Iberian Pig and Iberian x Duroc crossbreeding) labelled as "Iberian" into the NQS, due to the importance of these muscles in the Spanish fresh meat market, being actually the most expensive meat cuts of Iberian pork for fresh consumption.

The aim of this study was to compare the meat quality parameters between Iberian and Iberian x Duroc crossbreeding pork, currently undifferentiated in the Spanish market.

II – Materials and methods

1. Preliminary genetic analysis

In order to verify the racial origin of the selected pigs (Iberian and Iberian x Duroc pigs), a preliminary genetic study was conducted. This study was carried out on 25 animals, 15 assigned to Iberian Pig breed and 10 assigned to the Iberian x Duroc crossbreeding. All the animals were genotyped for several SNP of the MC1R and IGF2 genes using RT-PCR. These genes, following the methodology developed by the MERAGEM research group, can be used to differentiate Iberian pig breed from other breeds such as Duroc breed and Iberian x Duroc crossbreeding. In fact, this methodology is officially used by AECERIBER to ensure racial purity of the boars and sows registered in the Studbook, through an agreement with MERAGEM research group.

2. Animal management

Twenty-five castrated male pigs were used for this meat quality study, 15 from Iberian breed and 10 from Iberian x Duroc crossbreeding. All pigs were reared under regular semi-extensive management. Iberian and crossbred piglets were weaned at 49-56 days and fattening started at an age of about 12-13 weeks.

3. Sampling, carcass and meat quality analysis

The pigs were slaughtered when they reached the commercial live weight (150-170 kg; 10-12 months of age), and they were stunned according to the specifications outlined in the Spanish legislation. All measures (pH, weight percentages of moisture, ash, fat and protein, water holding capacity, Warner Bratzler shear force, muscle brightness and colour indices, concentration of myoglobin, and total fatty acids) were determined using standard methods.

4. Statistical analysis

Meat quality data were analyzed with the Statistica 7.0 for Windows statistical package (StatSoft, 2007). A general lineal model was used to determinate the significance of the effects of the different racial origins on meat quality traits. Carcass weight was fitted as a lineal covariate.

III – Results and discussion

1. Genetic analysis

Regarding the genotypes obtained from the study of the DNA molecular markers for the two analyzed genes (MC1R and IGF2) in the sampled animals, we must note that all animals preliminarily assigned to the Iberian pig breed showed the expected characteristic genotypes. On the other hand, all animals preliminarily considered Iberian x Duroc crossbreeding at 50 %, showed heterozygous genotypes (with a characteristic allele from Iberian Pig Breed and the other allele from Duroc Breed, for the two analyzed genes). Therefore, these results confirm a correct sampling of the selected animals.

2. Meat quality analysis

No differences between genetic groups (P>0.05) were observed for pH 24 h in analyzed carcass. The values ranged from 6.09 to 6.14 and from 6.07 to 6.11, in tenderloins and *serratus ventralis* muscles, respectively. These values were similar to those observed for tenderloin by Morcuende *et al.* (2007) and for *semimembranosus* muscle by Serrano *et al*. (2008).

Chemical composition and texture traits of tenderloins and *serratus ventralis* muscles from Iberian and crossbred pigs (Iberian x Duroc crossbreeding) are shown in Table 1. The shown values are similar to those reported by other authors for *longissimus dorsi* muscle of Iberian pigs (Estévez *et al.,* 2003; Cava *et al.,* 2004). Significant differences between Iberian and crossbred pigs were observed for protein, intramuscular fat, moisture and ash contents, as well as for water holding capacity in tenderloins. However, differences between the two analyzed genetic groups were observed only for protein content in *serratus ventralis* muscle. No differences (P > 0.05) between Iberian and crossbred pigs were found for shear force in neither of the two studied muscles.

Table 1. Proximate composition and texture traits of tenderloins and *serratus ventralis* muscles from Iberian and crossbred Iberian pigs

	Tenderloin			*Serratus ventralis*		
	Iberian	Crossbreed	Sig.	Iberian	Crossbreed	Sig.
Protein (%)	21.95 ± 0.366	19.84 ± 0.457	**	19.54 ± 0.508	17.98 ± 0.635	*
IMF (%)	4.41 ± 0.328	3.93 ± 0.446	*	4.19 ± 0.341	4.47 ± 0.401	ns
Moisture (%)	72.95 ± 0.354	75.19 ± 0.442	**	71.40 ± 0.390	71.37 ± 0.488	ns
Ash (%)	1.33 ± 0.037	1.06 ± 0.046	***	1.22 ± 0.032	1.18 ± 0.040	ns
WHC (%)	16.23 ± 0.728	12.24 ± 0.909	**	11.95 ± 0.777	12.90 ± 0.971	ns
WBSF (kg/cm^2)	4.51 ± 0.183	5.02 ± 0.229	ns	5.51 ± 0.358	4.76 ± 0.447	ns

IMF: intramuscular fat; WHC: water holding capacity; WBSF: Warner-Brätzler shear force.
Sig.: significant differences (ns: $P \geq 0.05$; *: $P < 0.05$; **: $P < 0.01$; ***: $P < 0.001$).

Physicochemical colour parameters (brightness, colour indices and myoglobin content) between Iberian and crossbred pigs of tenderloin and *serratus ventralis* muscle are shown in Table 2.

Table 2. Physicochemical colour parameters of tenderloins and *serratus ventralis* muscles from Iberian and crossbred Iberian pigs

	Tenderloin			*Serratus ventralis*		
	Iberian	**Crossbreed**	**Sig.**	**Iberian**	**Crossbreed**	**Sig.**
L^*	30.91 ± 0.618	38.39 ± 0.772	***	33.64 ± 0.460	38.62 ± 0.575	***
a^*	14.48 ± 0.481	10.29 ± 0.601	***	14.18 ± 0.378	10.37 ± 0.473	***
b^*	12.82 ± 0.296	6.70 ± 0.370	***	12.67 ± 0.385	8.64 ± 0.481	***
Mb (mg/100g)	4.92 ± 0.152	3.25 ± 0.190	***	5.29 ± 0.126	3.84 ± 0.158	***

L^*, a^* and b^*: muscle brightness and colour indices (CIE, 1976); Mb: myoglobin.
Sig.: significant differences (***:P<0.001).

The most significant differences between Iberian and crossbred pigs were found in these meat quality parameters. The redness value (a^*), as well as the myoglobin content, were higher ($P < 0.001$) in muscles from Iberian pigs, while the brightness (L^*) was lower ($P < 0.001$) in muscles from Iberian than in muscles from crossbred pigs. These data are in accordance with previous studies (Fernández *et al.,* 1999; Estévez, *et al.,* 2003). Iberian pigs have been reported to have higher concentration of oxidative fibres in muscles than less rustic breeds such as Duroc (Serrano *et al.,* 2008). Since muscles from Iberian pigs have more heme pigments (and therefore more iron) than muscles from crossbred pigs, muscles from Iberian pigs have higher redness value and less brightness than muscles from crossbred pigs. These result in an intense dark red colour.

Fat quality parameters, such as intramuscular fat content, marbling and lipid composition, are the main factors affecting consumer acceptability of Iberian fresh meat (Ruiz *et al.,* 2002). Moreover, the study of lipid composition of fat in fresh meat has acquired much importance in recent years mainly due to its correlation with cardiovascular diseases.

Relative percentages of individual fatty acids in intramuscular fat of tenderloins and *serratus ventralis* muscles (results not shown) revealed that the oleic acid (C18:1 n-9) was the most common fatty acid for both analyzed muscles in all sampled animals, followed by the palmitic (C16:0), stearic (C18:0) and linoleic (C18:2 n-6) acids. In general, no significant differences ($P \geq 0.05$) between Iberian and crossbred pigs were found for those majoritary fatty acids in each analyzed muscle, with the exception of palmitic acid ($P < 0.001$) in tenderloin. However, significant differences were found between Iberian and crossbred pigs for smaller fatty acids in intramuscular fat from both analyzed muscles, which may have nutritional and organoleptic influences.

Due to the high variability in the results, no significant differences ($P \geq 0.05$) were found between Iberian and crossbred pigs for the fatty acid main indices in *serratus ventralis* muscle (Table 3). However, compared to crossbred pigs, Iberian pigs had higher PUFA ($P < 0.05$), PUFA/SFA ($P < 0.05$) and UFA/SFA ($P < 0.01$) levels, and lower SFA ($P < 0.01$) values of the intramuscular fat of the tenderloins. In fact, PUFA/SFA ratio of intramuscular fat of tenderloins from Iberian pigs was the only one above 0.4, the international health recommendation (Department of Health, 1994).

Table 3. Composition of fatty acid indices of intramuscular fat of tenderloins and *serratus ventralis* muscles from Iberian and crossbred Iberian pigs

	Tenderloin			*Serratus ventralis*		
	Iberian	**Crossbreed**	**Sig.**	**Iberian**	**Crossbreed**	**Sig.**
SFA	38.24 ± 0.550	41.07 ± 0.687	**	38.35 ± 0.476	38.83 ± 0.594	ns
MUFA	43.38 ± 0.679	44.55 ± 0.849	ns	50.14 ± 0.542	49.28 ± 0.677	ns
PUFA	18.38 ± 0.929	14.38 ± 1.161	*	11.50 ± 0.501	11.89 ± 0.625	ns
PUFA/SFA	0.48 ± 0.029	0.35 ± 0.036	*	0.30 ± 0.015	0.31 ± 0.018	ns
UFA/SFA	1.62 ± 0.036	1.44 ± 0.045	**	1.61 ± 0.033	1.58 ± 0.041	ns
n-6/n-3	10.15 ± 0.458	10.26 ± 0.573	ns	8.10 ± 0.447	8.58 ± 0.559	ns

SFA: saturated fatty acids; MUFA: monounsaturated fatty acids; PUFA: polyunsaturated fatty acids; UFA: unsaturated fatty acids; n-6/n-3: omega-6 and omega-3 fatty acid ratios (Juárez, 2009). Sig.: significant differences (ns: $P \geq 0.05$; *: $P < 0.05$; **: $P < 0.01$).

IV – Conclusions

Tenderloins from Iberian pigs have different characteristics from that of crossbred pigs, commonly found in the Spanish meat market of Iberian products. These significant differences between Iberian and crossbred pigs in tenderloins would support a better labelling that explicitly differentiate the products from the two genetic groups. However, no meat quality differences between Iberian and crossbred pigs were found in *serratus ventralis* muscles, due to the heterogeneous characteristics and the different metabolism of these muscles compared with the tenderloins. Therefore, according to physicochemical meat quality parameters from Iberian and crossbred pig products, it appears that differences affect certain meat cuts and not the complete carcass. It would be interesting to carry out a study of the complete carcass on a higher number of animals to obtain reliable conclusions.

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Study of shelf life of liver pâté elaborated from Celta pig breed

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Abstract. In the present work, microbial spoilage, lipid oxidation, increase of non-heme iron content and colour changes occurring during refrigerated storage (75 days/4°C) of liver pâtés from Celta pigs were studied. Psychrotrophs and TVC increased during storage reached final values of 6.64 and 7.69 log cfu/g, respectively. *Brochothrix thermosphacta* was detected after 46 days of storage. During refrigerated storage L* values were maintained in a range of 63.8-65.4, whereas redness value decrease during the first point of sampling and after maintained a constant value close to 2.45. On the contrary yellowness value increased in the first 14 storage day, reached a value of 15.3, during the rest of storage period this value decrease until 14.4. Fe-heme content decrease during shelf life period from 17.30 to 13.5 mg Fe-heme/kg liver pâté, whereas Fe-total content presented a inverse relationship, because increase from 110 to 223 mg Fe-total/kg liver pâté. However, this high amount of Fe-total did not seem to be related to oxidative process, because even thought TBAR´S values increase during the firsts 60 days of display period, decreasing in the last sampling point, reaching a final values of 0.015 mg malonaldehyde/kg of liver pâté, this final value was indicate that liver pâté did not undergo important lipid oxidation.

Keywords. Pâté – Celta pig – Refrigeration – Oxidation stability – Non-heme iron.

Étude de la vie utile du pâté élaboré à partir du porc Celta

Résumé. *Dans ce travail, l'étude de divers paramètres a été menée sur du pâté élaboré à partir de porc Celta: il s'agit de la contamination microbienne, l'oxydation lipidique, l'augmentation du fer non héminique et les changements de coloration produits au cours de la réfrigération (75 jours à 4ºC). Les comptages de psychrotrophes et TVC ont augmenté de manière significative (P<0,001) au cours de l'étape de conservation atteignant respectivement des valeurs de 6,64 et 7,69 log ufc/g.* Brochothrix thermosphacta *fut détecté après 46 jours de conservation. Au cours de la conservation à froid, les valeurs de luminosité sont demeurées constantes à 63,8-65,4, tandis que les indices de rouge ont diminué durant le premier point d'échantillonnage, pour se maintenir constants par la suite à une valeur proche de 2,45. En revanche, l'indice de jaune a augmenté au cours des 14 premiers jours de conservation, atteignant une valeur de 15,3 et régressant à un niveau de 14,4 au cours du restant de la période de conservation. La teneur en fer héminique a diminué au cours de la période de vie utile passant de 17,3 à 13,5 mg de fer haem/kg de pâté, tandis que la teneur en fer total a adopté un comportement inverse puisqu'elle a augmenté de 110 à 223 mg Fe-total/kg pâté. Dans tous les cas, la présence de cette quantité de fer total ne semble pas avoir d'influence sur le processus d'oxydation lipidique, car bien que les valeurs de TBAR's aient augmenté au cours de la période de conservation, atteignant 0,015 mg de malonaldéhyde/kg de pâté, elles ne l'ont pas fait de manière assez significative pour pouvoir conclure à une importante oxydation lipidique.*

Mots-clés. *Pâté – Porc Celta – Réfrigération – Stabilité par oxydation – Couleur – Fer non héminique.*

I – Introduction

Apart from microbial spoilage, lipid oxidation is the major factor reducing quality and acceptability of meat and fat products (Morrissey *et al.*, 1998). Lipid oxidation is a complex process whereby polyunsaturated fatty acids are degraded via formation of free radicals, causing flavour, texture, colour and nutritional deterioration of foodstuffs (Gray, 1978). Non-heme iron (NHI) is considered the most important oxidation promoter in meat systems and, therefore, knowledge of the proportions of the chemical forms of iron is of great importance (Kanner *et al.*, 1991). An increase in the amount of NHI as a result of thermal processes on meat systems has been shown (Lombardi-Boccia, *et al.*, 2002). Miller *et al.* (1994), suggested cooking is not as important as the subsequent refrigerated storage of cooked meats for the release of NHI from myoglobin. The increase of NHI in meats and fish is considered to be a reflection of the decrease of heme iron (HI) as a consequence of the breakdown of the heme molecule during cooking or storage (Gómez-Basauri and Regenstein, 1992a; Gómez-Basauri and Regenstein, 1992b) and this has been linked to the oxidative deterioration of the porphyrin ring of myoglobin (Schricker and Miller, 1983).

The colour of meat products is another important quality attribute that influences consumer acceptance, and a brown-gray colour is preferred for cooked products (Cornforth, 1994). Colour changes in cooked products during refrigerated storage have been linked to oxidation phenomena, and several factors such as the characteristics and amount of fat, the packaging method and the presence of antioxidants have been reported as being influential (Jo and Ahn, 1999).

Liver pâté is a traditional product for which there has been an increasing demand by European consumers in the last 15 years (Rosmini *et al.*, 1996). Liver pâtés contain high amounts of fat and iron, and therefore, oxidative deterioration of liver pâtés during refrigeration is expected. The differences between pâtés from Celta and white pigs in terms of their fatty acid composition and antioxidative status are expected to influence their susceptibility to oxidative deterioration during refrigerated storage.

The aim of the present work was to study the microbial changes of liver pâtés from Celta pigs during refrigerated storage as assessed by lipid oxidation, increase in the amount of NHI and colour deterioration.

II – Materials and methods

1. Experimental design

For the manufacture of the pâtés (1.5 kg), muscles and adipose tissues from Celta pigs were used. In the recipe the ingredients were as follows per 100 g of product: 27 g liver, 26 g adipose tissue, 26 g muscle, 16 g chestnut, 2 g sodium caseinate, 2 g sodium chloride. The procedure for the manufacture of the pâtés has been described by Estévez *et al.* (2004). Liver pâtés were packed in glass containers prior to thermal treatment (80°C/30′). After the containers were allowed to cool at room temperature, they were stored in the dark at 4°C for 75 days from the day of the manufacture (day 0). Liver pâtés were analysed at days 0, 14, 48, 60 and 75 for lipid oxidation, concentration of NHI, instrumental colour and microbial counts. After each of the refrigeration stages, instrumental colour was measured on the surface of the pâtés and then they were stored at −80°C until the analytical measurements were carried out.

2. Analytical methods

A. Microbial analyses

In each liver pâtés unit, after aseptically removing and discarding the outer plastic, 10 g of the product were aseptically taken and homogenized with 90 ml of sterile 0.1% peptone water also containing 0.85% NaCl and 1% Tween 80 as emulsifier, at 40-45°C for 2 min in a Masticator blender (IUL Instruments, Barcelona, Spain), thus making a 1/10 dilution. Successive decimal dilutions were prepared by mixing 1 ml of the previous dilution with 9 ml sterile 0.1% peptone water.

Phychrotroph microflora was enumerated in Standard Plate Count Agar (PCA) agar (Merck), after incubation at 7°C for 10 d; *Enterobacteriaceae* in violet red bile dextrose (VRBD) agar

(Merck) after incubation at 37°C for 24 h; *Staphylococcus aureus* in Baird Parker agar (Merck) + Egg Yolk Tellurite Emulsion (Biokar Diagnostics) incubated at 37°C for 24 h and Sulfite reducing clostridia in Perfringens Selective Agar (SPS) agar (Merck) after incubation at 44ºC for 24 h. Presence or absence of *Salmonella* was investigated by Enzyme Linked Fluorescent Assay (ELFA), VIDAS®-SLM protocol was carried out according to the procedures recommended by the manufacturer. From each sample and on each culture medium, 1 ml of each dilution was inoculated in duplicate on plates and mixed before solidification. Plates of VRBD agar were covered with a layer of the same culture medium before incubation. After incubation, plates with 30-300 colonies were counted.

B. Iron analysis

HI was measured according to the methodology of Hornsey (1956) with the next expressions (Merck, 1989):Hematin (µg hematin/ g muscle) = Absorbance × 342.44 and HI (mg/100 g meat) = (Hematin x 8.82)/100. Total iron (TI) content was measured following the methodology proposed by Lorenzo *et al.* (2003) and NHI was calculated as difference between TI and HI content.

C. Colour measurement

A portable colorimeter (Konica Minolta CR-400 Osaka, Japan) was used to measure liver pâtés colour in the CIELAB space (CIE 1978). (lightness, L*; redness, a*; yellowness, b*)

D. Measurement of TBARs

Lipid stability was evaluated in the liver pâtés using the method proposed by Vyncke (1975) with the modification that samples were incubated at 96 ºC in a forced oven (Memmert UFP600, Germany, Schawabach). Results are expressed as (mg malonaldehyde / kg of fresh meat).

III – Results and discussion

Figure 1 shows microbial spoilage evolution of TVC, psychrotrophs, LAB, enterobacteriaces and *Brochothrix thermosphacta.* All microbial populations increased during storage at 4ºC, reached final values of 7.69, 6.64, 6.74, 5.54 and 3.6 log cfu/g, respectively. *Brochothrix thermosphacta* was detected after 46 days of storage.

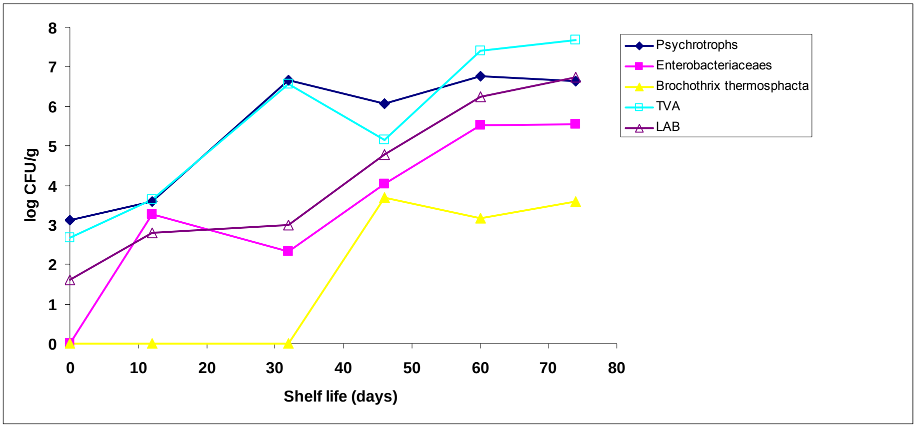

Fig. 1. Evolution of microbial groups (TVC, psychrotrophs, LAB, enterobacteriaces and *Brochothrix thermosphacta*) on liver pâté from Celta pig under refrigeration storage.

Table 1 shows colour characteristics evolution of liver pâté. During refrigerated storage L* values was maintained in a range of 63.8-65.4, whereas redness value decrease during the first point of sampling and after maintained a constant value close to 2.45. On the contrary yellowness value increased in the first 14 storage day, reached a value of 15.3, during the rest of storage period this value decrease until 14.4. Compared to pâtés from Iberian pigs of Estévez and Cava (2004) pâtés from our work presented a darker colour (64.52 *vs* 65.48) with less redness (8.43 *vs* 2.68) and higher yellowness (12.14 *vs* 14.39) during 90 days of storage in similar conditions. Obviously differences in the recipe, colour characterises of the meat and adipose tissue, feeding regime and breed explain these differences.

Table 1. Evolution of colour parameters, ashes, total iron, non-heme and heme iron and lipid oxidation of liver pâté from Celta pig under refrigeration storage

Colour characteristics	Shelf life (days)				
	0	14	48	60	75
Luminosity (L*)	63.84	63.68	64.15	63.93	65.48
Redness (a*)	3.85	2.60	2.40	2.21	2.68
Yellownes (b*)	12.48	15.33	14.55	14.78	14.39
Ashes-Fe content					
Ashes (%)	1.76	2.04	2.12	2.06	2.22
TI (ppm)	110.8	177.0	201.0	178.1	223.0
NHI (ppm)	93.5	164.7	188.8	163.6	209.5
HI (ppm)	17.3	12.3	12.2	14.4	13.5
Lipid oxidation					
TBARS (mg MDA/kg pâté)	0.149	0.136	0.179	0.318	0.015

NHI content increased during refrigerated storage, from 93 to 209 mg/kg pate from day 0 to day 75, whereas HI content decrease during shelf life period from 17.30 to 13.5 mg /kg liver pâté and TI content presented a inverse relationship, because increase from 110 to 223 mg TI/kg liver pâté (Table 1). Results suggest that some disruption of the porphyrin ring could have occurred during storage that led to the release of iron. For Gómez-Basauri and Regenstein (1992a) and Miller *et al.* (1994) the increase of NHI during refrigeration of meat is a reflection of the degradation of HI. Damage in the porphyrin ring during cooking or storage has been suggested to cause the breakdown of heme molecule and the release of iron from globin (Gómez-Basauri and Regenstein, 1992a). The degradation of HI would reduce the nutritional value of the pâtés in terms of bioavailability of iron, since HI is more available than NHI (Hunt and Roughead, 2000).

However, this high amount of TI did not seem to be related to lipid oxidative process, because TBARS of liver pâtés increasing during 60 days of refrigerated storage, decreasing in the last 15 days of shelf life to a close value to 0. This final value indicates that liver pâté did not undergo important lipid oxidation or also it could be explained by disappearance of primary oxidation products. Secondary peroxidation products such peroxides, could be present in the liver pâté but was not measured.

IV – Conclusions

According to this study, lipid oxidation, and the increase of NHI during refrigerated storage of liver pâtés could not be closely related. Colour changes seem not to be linked to oxidative processes and microbial counts. This previous results represent a starting point for the promotion elaborated meat products from this endangered pig breed.

Acknowledgements

Authors are grateful to FEADER (Project 2008-15) for the financial support. Special thanks to Porco Celta Fonsagrada, S.L. for liver pâtés from Celta pig samples supplied for this research.

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Manufactured of "chanfaina", from Celta pig breed. Study of shelf life vacuum packaging

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Abstract. *Chanfaina* is a dry-fermented sausage abundantly produced and consumed in Galicia (NW Spain) elaborated from Celta pig breed. Traditionally, this product is distributed and sold without packaging in the local market. To extend its shelf life and expand the market, some manufacturers have begun to implement vacuum packaging. Total viable count (TVC), psychrotrophs, lactic acid bacteria (LAB), pseudomonas, enterobacteria, moulds and yeasts, *Staphylococcus aureus*, sulfite reducing clostridia, pH and TBAR´S value were analysed during storage at 4 °C. Sulfite-reducing clostridia and enterobacteria were not detected in any sample. Psychrotrophs were the predominant microorganisms reaching a population higher than 8.9 log cfu/g after 60 days of storage. At the same time, LAB becomes the predominant species during storage. The rest of the microbiota did not grow during storage. On the other hand, a lightly increase in pH was noticed during storage. Samples stored in vacuum package remained stable during the whole display period and no significant differences (P<0.05) were observed in TBAR´S values.

Keywords: *Chanfaina* – Celta pig breed – Vacuum packaging – Dry-fermented sausages.

Élaboration de "chanfaina" à partir de porc Celta. Étude de la vie utile après emballage sous vide

***Résumé.** La Chanfaina est une saucisse sèche produite à partir de porc Celta en Galice (nord-ouest de l'Espagne) où elle y est hautement consommée. Ce produit est traditionnellement distribué et vendu sans emballage dans les marchés locaux. Dans le but d'accroître sa vie utile et d'élargir son rayon de vente, certains fabricants ont commencé à mettre en œuvre un emballage sous vide. Les comptages de mésophiles aérobies totaux, psychrotrophes, bactéries acido-lactiques, pseudomonas, entérobactéries, moisissures et levures,* Staphylococcus aureus*, clostridia sulfito-réducteurs, pH et TBAR´S, ont été déterminés durant la conservation à 4ºC. Aucune trace de clostridia sulfito-réducteurs et d'entérobactéries n'a pu être observée au sein des échantillons. Les micro-organismes prédominants furent les psychrotrophes, avec des valeurs atteignant 8,8 log ufc/g après 60 jours de conservation. Dans le même temps, LAB est apparue comme l'espèce majeure formée durant la conservation. Une légère augmentation du pH fut également observée. Les échantillons conservés sous vide sont demeurés stables durant toute l'étape de conservation et aucune variation significative des valeurs de TBAR'S (P < 0,05) n'a pu être mise en évidence.*

***Mots-clés.** Chanfaina – Porc Celta – Emballage sous vide – Saucisse sèche.*

I – Introduction

The Celta was the typical breed of pig raised on farms in Galicia (northwest Spain) until the middle of the 20th century, at which time it suffered an important recession in members due to the introduction of improved breeds and their crossbreds. This breed is highly appreciated by consumers because of the succulent meat that results from the profuse infiltration of fat into the lean meat (Franco *et al.,* 2006).

"Chanfaina" is a fermented and dried-ripened sausage abundantly produced from raw Celta pork in Galicia (NW of Spain). For the manufacture of chanfaina, low-quality pork (lean, bacon, lung, heart, jowls) is minced and salt, sweet and spicy paprika, garlic and onion are added. The resulting mass is left standing for at least 24 h and then, it is stuffed into pork rectum tripe in units of 20-25 cm length. After stuffing, it undergoes a smoking-heating process for 8-10 days and then a drying-ripening process for 15 days.

Oxidation of the lipid fraction is one of the major causes of quality decrease during the shelf-life of sausages. The extent of the overall lipid degradation process may be affected by various factors related to: (i) the storage conditions (García-Esteban *et al.,* 2004; Papadima, and Bloukas, 1999; Zanardi *et al.,* 2002); (ii) the processing technology (Gray *et al*., 1996; Salgado *et al.,* 2005); (iii) the additives used in the dough formulation (Kanner, 1994; Skibsted, 1992); and (iv) the polyunsaturated fatty acids content of the lipid fraction.

The aim of this study was to evaluate the microbial changes of chanfaina from Celta pigs during refrigerated storage as assessed by lipid oxidation.

II – Materials and methods

1. Samples

Twenty units of chanfaina were manufactured by Porco Celta Fonsagrada, SL following the procedure described in introduction section. Samples were vacuum packed prior to thermal treatment (100ºC/15 min). After the samples were allowed to cool at room temperature, they were stored in the dark at 4°C for 180 days.

2. Microbial analyses

In each chanfaina unit, after aseptically removing and discarding the outer plastic, 10 g of the product were aseptically taken and homogenized with 90 ml of sterile 0.1% peptone water also containing 0.85% NaCl and 1% Tween 80 as emulsifier, at 40-45 °C for 2 min in a Masticator blender (IUL Instruments, Barcelona, Spain), thus making a 1/10 dilution. Successive decimal dilutions were prepared by mixing 1 ml of the previous dilution with 9 ml sterile 0.1% peptone water.

Phychrotroph microflora was enumerated in Standard Plate Count Agar (PCA) agar (Merck), after incubation at 7°C for 10 d; *Enterobacteriaceae* in violet red bile dextrose (VRBD) agar (Merck) after incubation at 37°C for 24 h; *Staphylococcus aureus* in Baird Parker agar (Merck) + Egg Yolk Tellurite Emulsion (Biokar Diagnostics) incubated at 37°C for 24 h and Sulfite reducing clostridia in Perfringens Selective Agar (SPS) agar (Merck) after incubation at 44ºC for 24 h. Presence or absence of *Salmonella* was investigated by Enzyme Linked Fluorescent Assay (ELFA), VIDAS®-SLM protocol was carried out according to the procedures recommended by the manufacturer. From each sample and on each culture medium, 1 ml of each dilution was inoculated in duplicate on plates and mixed before solidification. Plates of VRBD agar were covered with a layer of the same culture medium before incubation. After incubation, plates with 30-300 colonies were counted.

3. pH measurement

pH was measured by blending 25 g of product with 225 ml of distilled water for 2 min. A digital pH-meter (Hanna HI 99163, Spain) was used for the measurement.

4. Measurement of TBARs

Lipid stability was evaluated in the steaks using a small 2 g portion. Lipid oxidation, measured by aldehydes generated in the process of polyunsaturated fatty acid oxidation, was determined by measuring 2-thiobarbituric acid reactive substances (TBARs) using the method proposed by Vyncke (1975) with the modification that samples were incubated at 96ºC in a forced oven. Results are expressed as (mg malonaldehyde / kg of fresh meat).

III – Results and discussion

1. Microbial characteristics

TVC, psychrotrophs, LAB, moulds and yeasts and pseudomonads are showed in Fig 1. Sulfite-reducing clostridia and enterobacteria were not detected in any of the samples analysed. The aerobic mesophilic bacteria, psychrotrophs and lactic acid bacteria counts were over 6.4, 7.8 and 8.1 log cfu/g, respectively immediately after packaging, while pseudomonads and moulds and yeasts were below over 3 log cfu/g.

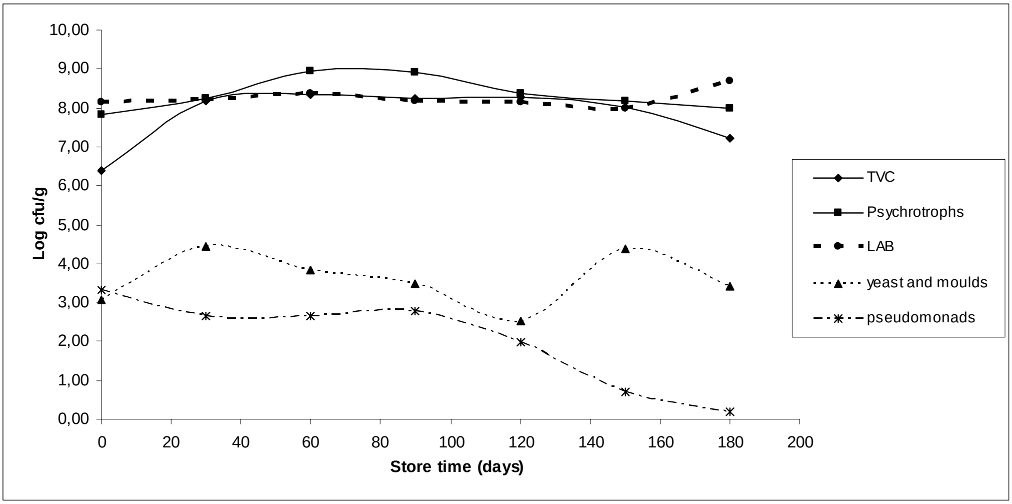

Fig. 1. Evolution of TVC, psychrotrophs, LAB, moulds and yeasts and pseudomonads in chanfaina from Celta pigs under refrigerated storage. Means with different letter in the figure show significant differences (P<0.05; Duncan test) for the effect store time.

2. pH changes

Initial pH values were below 5 for all samples analysed (Fig. 2). A non-significant increase (P>0.05) was observed during all the period of storage. However, it can show a lightly increased in pH values under refrigerated storage reached a mean final pH value of 5.18.

3. Lipid stability

The level of lipid oxidation of chanfaina was estimated on base of the amount of 2-thiobarbituric acid reactive substances (TBAR´s values) (Fig. 2). All samples started with low values of about 0.4 mg MDA/kg in fresh meat. Exclusion of the oxygen content in the vacuum package limited oxidation and thus resulted in lower TBAR´s values for these *chanfaina* samples. Samples stored in vacuum package remained stable during the whole display period and no significant differences (P<0.05) were observed. This outcome was not surprising, as the meat storage conditions during the display period in a vacuum environment protect the meat from oxygen.

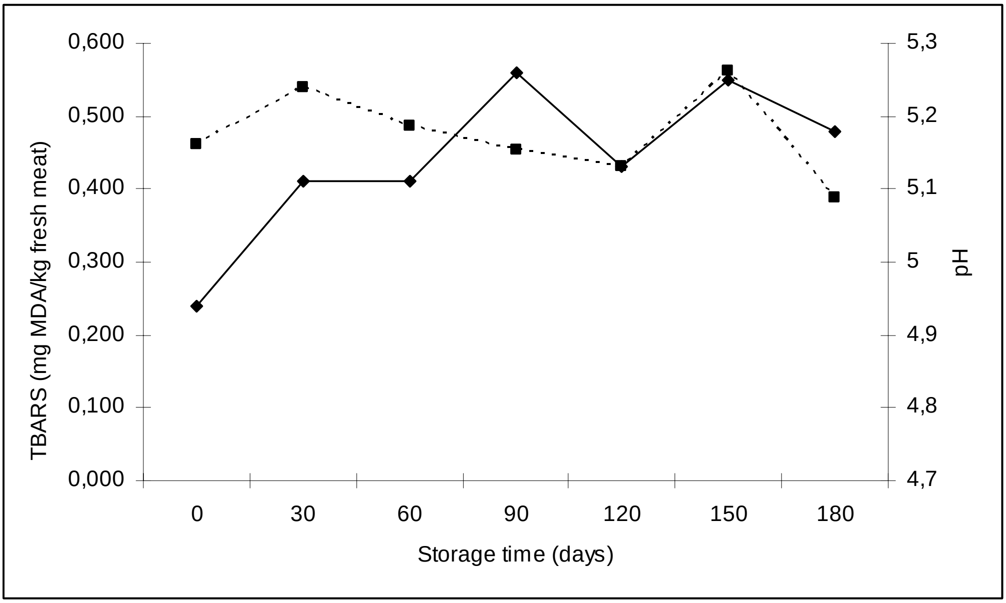

Fig. 2. Evolution of pH and TBAR´S values in chanfaina from Celta pigs under refrigerated storage.

Acknowledgements

Authors are grateful to FEADER (Project 2008-15) for the financial support. Special thanks to Porco Celta Fonsagrada, S.L. for chanfaina from Celta pig samples supplied for this research.

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Manufactured of "Mestura cocida" from Celta pig breed. Study of shelf life vacuum packaging

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Abstract. *"Mestura cocida"* is a typical product manufacture and consumed in Galicia (NW Spain) elaborated with dry-cured "lacón", "Galician chorizo" and salted head from Celta pig breed. The elaboration process is strictly controlled to achieve a satisfactory shelf life, so a high quality finally-consumed product is obtained. Physical, chemical, enzymic and microbiological test are performed to ensure this quality. Psychrotrophs, Enterobacteriaceae, *Staphylococcus aureus*, *Salmonella*, sulfite reducing clostridia and TBAR´S value were analysed during storage at 4°C. Psychrotrophs and Enterobacteriaceae showed a increase during storage observed final values 7.88 and 5.12 log cfu/g, respectively after 90 days of storage. *Staphylococcus aureus* and *Salmonella* have not been found in any samples. On the other hand, TBAR´S values increased) during the whole display period reaching a final values of 11.8 mg malonaldehyde / kg of "Mestura cocida".

Keywords. Mestura cocida – Celta pig breed – Vacuum packaging.

Élaboration de "mestura cocida" à partir de porc Celta. Étude de la vie utile après emballage sous vide

***Résumé.** La "Mestura cocida" est un produit typique produit et consommé en Galice (nord-ouest de l'Espagne) qui est fabriqué à partir de "lacón", de "chorizo galicien" et de tête de porc Celta salée. Afin d'établir la vie utile d'une denrée alimentaire et de s'assurer de sa qualité finale, il existe diverses mesures de type physique, chimique, enzymatique et microbiologique. Les comptages de psychrotrophes, entérobactéries,* Staphylococcus aureus*,* Salmonella*, clostridia sulfito-réducteurs et les valeurs de TBAR´S, ont été déterminés durant la conservation à 4 ºC. Après 90 jours d'exposition, une augmentation de psychrotrophes et d'entérobactéries fut observée durant le temps de vie utile, atteignant respectivement des valeurs de 7,88 et 5,12 log ufc/g. Aucune trace de* Staphylococcus aureus *et de* Salmonella *n'a pu être détectée au sein des échantillons. Par ailleurs, une croissance importante des valeurs de TBAR´S fut également observée au cours de la période de conservation, pour atteindre des valeurs finales de 11,8 malonaldéhyde/kg de mestura cocida.*

***Mots-clés.** Mestura cocida – Porc Celta – Emballage sous vide.*

I – Introduction

"Mestura cocida" is a typical product manufacture and consumed in Galicia (Nw Spain) elaborated mainly with dry-cured "lacón", "Galician chorizo" and salted head from Celta pig breed. Dry cured lacón and Galician chorizo are traditional raw-cured meat products made in the northwest of Spain. Dry-cured lacón is elaborated from the foreleg of the pig, using similar manufacturing processes to those used in the production of dry-cured ham, Galician chorizo can be defined as the mixture of minced pork and pork fat, adition of salt, paprika, other spices and additives, mixed and inserted into natural or artificial casings, which undergo a drying-ripening process. The Celta was the typical breed of pig raised on farms in Galicia until the middle of the 20th century. This breed is highly appreciated by consumers because of the succulent meat that results from the profuse infiltration of fat into the lean meat.

The elaboration process of this product is controlled to achieve a satisfactory shelf life, so a high quality finally consumed product is obtained. This product is subjected to a pasteurisation process and vacuum-packaging. For this reason these cooked meat products are often post-contaminated because of a packaging and/or slicing step after pasteurisation process. After cooking, the normal flora of the product, is too low to protect the products against the growth of Gram-negative microorganisms (Kotzekidou and Bloukas, 1996). The bacterial flora is gradually selected towards a CO_2-tolerant but a slowly growing one (Borch *et al*., 1996). Psychrotrophic lactic acid bacteria are responsible for the spoilage of cooked meat products packed in oxygen-free atmospheres (Debevere, 1989; Borch *et al*., 1996). Enterobacteriaceae, *Staphylococcus aureus*, *Salmonella* and sulfite reducing clostridia are indicative microorganisms of the hygienic quality of the product and their count can give us an idea from the possible later contamination to the thermal treatment as well as of the effectiveness of the same one.

Oxidation of the lipid fraction is one of the major causes of quality decrease during the shelf life of mestura cocida. In particular, the deterioration involves modifications of the organoleptic characteristics, the development of unpleasant odours and tastes and a decrease in the nutritional value of the product due to a lowering of the polyunsaturated fatty acid content, whose beneficial effect on consumers health is well-known (Alexander, 1978; Rose and Connolly, 1999; Berra *et al*., 2005). Although vacuum packaging can protect meat from contamination and increase the shelf life of the product, tha anaerobic conditions may affect the quality. Many studies have revealed a change in the prevailing microflora in vacuum-packed meat products compared to that prevailing before storage (Björkroth *et al*., 1998; Samelis *et al*., 2000). This microbiological change could result in some modifications of the sensory properties and it could affect the nutritional value and the chemical compositions of products.

The purpose of the present work was, to assess the quality, during of shelf life, of vacuum-packaging mestura cocida elaborated from Celta pig breed. Physical, chemical and microbiological test are performed to ensure this quality.

II – Material and methods

1. Samples

Fifteen units of mestura cocida were manufactured by Porco Celta Fonsagrada, SL. For the manufacture of this product muscles and adipose tissues from Celta pigs were used (40% dry-cured "lacón", 40% salted head and 20% "Galician chorizo"). Samples were vacuum packed prior to thermal treatment (80 ºC/30 min). After the samples were cooled at room temperature, and stored at 4 °C for 90 days from the day of the manufacture (day 0). "Mestura cocida" samples were analysed at days 0, 30, 45, 60, 75 and 90 for lipid oxidation and microbial counts. In every sample point two units of "mestura cocida" were analyzed.

2. Microbiological analysis

In each mestura cocida unit, after aseptically removing and discarding the outer plastic, 10 g of the product were aseptically taken and homogenized with 90 ml of sterile 0.1% peptone water also containing 0.85% NaCl and 1% Tween 80 as emulsifier, at 40-45°C for 2 min in a Masticator blender (IUL Instruments, Barcelona, Spain), thus making a 1/10 dilution. Successive decimal dilutions were prepared by mixing 1 ml of the previous dilution with 9 ml sterile 0.1% peptone water.

Phychrotroph microflora was enumerated in Standard Plate Count Agar (PCA) agar (Merck), after incubation at 7°C for 10 d; *Enterobacteriaceae* in violet red bile dextrose (VRBD) agar (Merck) after incubation at 37°C for 24 h; *Staphylococcus aureus* in Baird Parker agar (Merck) + Egg Yolk Tellurite Emulsion (Biokar Diagnostics) incubated at 37°C for 24 h and Sulfite reducing clostridia in Perfringens Selective Agar (SPS) agar (Merck) after incubation at 44ºC for 24 h. Presence or absence of *Salmonella* was investigated by Enzyme Linked Fluorescent Assay

(ELFA), VIDAS®-SLM protocol was carried out according to the procedures recommended by the manufacturer. From each sample and on each culture medium, 1 ml of each dilution was inoculated in duplicate on plates and mixed before solidification. Plates of VRBD agar were covered with a layer of the same culture medium before incubation. After incubation, plates with 30-300 colonies were counted.

3. TBAR´s determination

Lipid stability was evaluated in the "Mestura cocida" using the method proposed by Vyncke (1975) with the modification that samples were incubated at 96 ºC in a forced oven (Memmert UFP600, Germany, Schawabach). Results are expressed as (mg malonaldehyde / kg of mestura cocida).

III – Results and discussion

Figure 1 shows the evolution of microbial counts in the mestura cocida during the shelf life vacuum packaging. Psychrotrophs bacterias and Enterobacteriaceae showed a increase during storage at 4ºC observed final values of 7.88 and 5.12 log cfu/g, respectively after 90 days of storage.

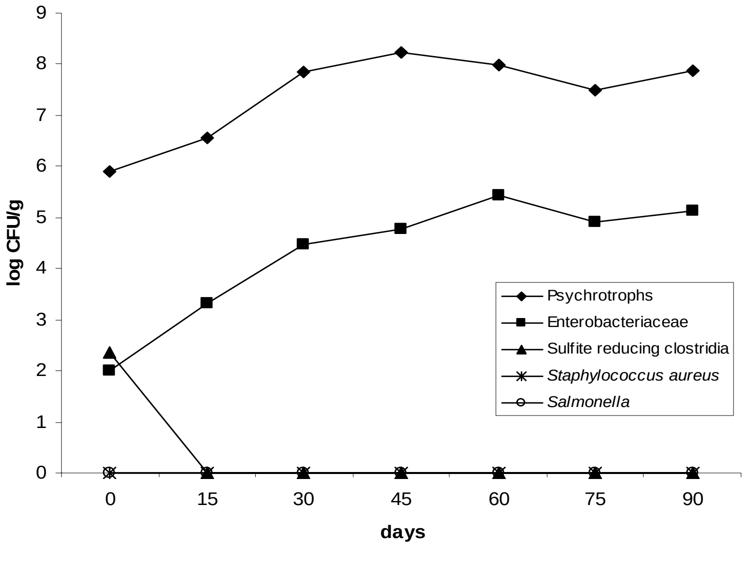

Fig. 1. Changes in log counts of the spoilage microflora in the mestura cocida under vacuum-packaging at 4ºC.

In general, mestura cocida samples showed high counts of Enterobacteriaceae and psychrotrophs bacterias. The Enterobacteriaceae populations indicate the hygienic quality of the product, and their presence can be related to contamination of faecal origin and high counts indicate poor hygienic practices or high contamination of the raw materials used in their manufacture.

Sulfite reducing clostridia only was found in the control sample. This fact could be related with the effectiveness of the thermal treatment. *Staphylococcus aureus* and *Salmonella* have not been found in any samples.

Figure 2 shows the development of lipid oxidation in the mestura cocida during the shelf life vacuum packaging. TBAR`S values increased during the whole display period reaching a final values of 11.8 mg malonaldehyde / kg of mestura cocida. Duration of display period affected the overall TBARS formation of the mestura cocida. The amounts of TBARS formed in the course of storage were higher the critical value of 3 mg/kg at which rancidity is detected (Wong *et al*., 1995).

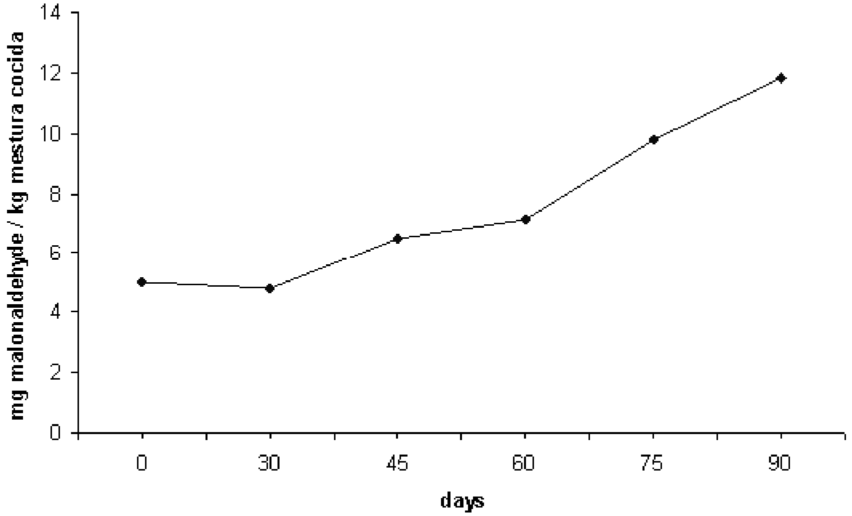

Fig. 2. Changes in TBAR's values in the mestura cocida during shelf life vacuum-packaging at 4 ºC.

Vacuum packaging changes the gaseous environment at the sample surface: respiration of microorganisms at the sample surface or the sample itself produces CO_2 and eventually the oxygen concentration within the pack falls below 1% while the CO_2 concentration rises to 20% or more (Eustace, 1981). The compositional changes of gas could have involved the control of oxygen-dependent microorganisms or oxidative degradation of meat in the bag.

IV – Conclusions

Mestura cocida showed high counts of Enterobacteriaceae and psychrotrophs bacterias. Sulfite reducing clostridia only was found in the control sample. *Staphylococcus aureus* and *Salmonella* have not been found in any samples. The amounts of TBARS formed in the course of storage were higher the critical value of 3 mg/kg at which rancidity is detected.

To extend the shelf life of this product would be advisable: reduction of microbial contamination during production, growth prevention of spoilage bacteria and application of decontamination procedures to product after packaging.

Acknowledgements

Authors are grateful to FEADER (Project 2008-15) for the financial support. Special thanks to Porco Celta Fonsagrada, S.L. for chanfaina from Celta pig samples supplied for this research.

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Consumers attitudes to Iberian pork meat

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Abstract. In the last years, Iberian pork consumption has been limited as a result of relations established between fat consumption and cardiovascular diseases. However, recent researches have allowed establishing healthy properties to the intake of these kind of meat, and as a consequence its sale has increased, competing successfully with other meats. The objective of this study was to determine the attitudes of consumers to Iberian pork meat. To assess consumers' behaviour to breed (Iberian and white pig) and anatomical origin of the pieces commercial ("*pluma*", "*presa*", "*secreto*") of Iberian pork meat, discriminate tests were used in order to detect differences. Besides, an affective test was carried out in order to know the consumers preference to feeding of the Iberian pigs ("*bellota*", "*cebo*"). The results obtained in the triangle tests showed that consumers were able to differentiate between Iberian and white pork loin. Also differences were found between different commercial cuts of Iberian pig. However, consumers didn't show preferences regarding feeding of the Iberian pigs.

Keywords. Sensory analysis – Meat – Iberian and white pig – Feeding system.

Attitudes des consommateurs vis-à-vis de la viande de porc Ibérique

Résumé. *Lors des dernières années, la consommation de porc Ibérique a été limitée en raison des relations établies entre la consommation de matières grasses et les maladies cardiovasculaires. Cependant, les récentes recherches ont permis d'attribuer des propriétés santé à la consommation de ces types de viande, ce qui a déterminé un développement de leur vente, concurrençant avantageusement d'autres viandes, grâce à leur qualité nutritive et leurs caractéristiques sensorielles. L'objectif de l´étude était de déterminer les attitudes des consommateurs vis-à-vis de la viande de porc Ibérique. Pour évaluer ce comportement, des tests discriminants ont été réalisés pour détecter des différences dues à la race (porc Ibérique et porc blanc) ou à l'origine anatomique de la pièce (*"pluma", "presa", "secreto"*) et des tests pour déterminer la préférence en ce qui concerne l'alimentation du porc Ibérique (*"bellota", "cebo"*). Les résultats obtenus dans le test triangulaire réalisé ont montré que les consommateurs étaient capables de différencier entre échine de porc Ibérique et de porc blanc. Ils ont aussi différencié entre différentes pièces de porc Ibérique. Cependant, ils n'ont montré aucune préférence en fonction de l'alimentation.*

Mots-clés. *Analyse sensorielle – Viande – Porc ibérique et porc blanc – Alimentation.*

I – Introduction

In the last years, Iberian pork consumption has been limited as a result of relations established between fat consumption and cardiovascular diseases. However, recent researches (García-Rebollo *et al*., 1998) have allowed establishing healthy properties to the intake of this kind of meat.

Iberian pork comes from genuinely bred Southwest Iberian Peninsula pigs traditionally fattened with acorns and pasture in an extensive production system. Most of Iberian pork is consumed as highly priced cured products. However, the importance of the consumption of fresh meat has recently increased, since consumers have become more concerned about questions such as ethical forms of animal production, animal welfare, traditional production or nutritional and sensory characteristics of the meat.

It is well know that breed, productive system and muscle type influence the fatty acids profiles and the sensory characteristic of fresh pork meat and meat products. However, the influence of these factors on eating quality of Iberian meat has not been evaluated.

The aim of this study was to determine if the sensory quality of culinary pork meat was affected by breed (Iberian and white pig), retail cuts ("*pluma*", "*presa*", "*secreto*"), and rearing system ("*bellota*", "*cebo*") using different tests with consumers.

II – Material and methods

1. Meat samples

Samples from heavy white pig (loin) were obtained from a regional abattoir (Segovia, Spain) the day after slaughter and samples from Iberian pig (loin, sirloin, "pluma", "presa", "secreto") were obtained from a local abattoir (Guijuelo, Spain) the day of slaughter. All the samples were transferred to Estación Tecnológica de la Carne, where they were vacuum-packed and frozen at -20ºC until further analyses.

2. Consumers

Consumers tests were performed during the "VI Jornadas del cerdo Ibérico y sus productos" (Salamanca-Spain, 2008). The tasters (n= 85) were mainly farmers, manufacturers and technicians associated with pig meat industry. Before tasting the samples, subjects were asked to specify their age, their gender and their frequency pork meat consumption on the evaluation sheet.

3. Analysis

Consumers were only informed that they would evaluate different pork samples.

A triangle test (ISO 4120:2004) was performed on loin (*longissimus lumborum* and *thoracis* muscle), to determine whether there was a difference between samples from Iberian and heavy white pig reared in confinement with a concentrate feed. The triangle test was carried out by the forced-choice option, in which the tasters must choose the sample that, in their opinion, is different. Besides, to complement the triangle test, tasters were asked to indicate their reasons for selecting one particular sample of the three used in the analysis.

A rank order test was carried out between "pluma" (rhomboid muscle chest), "presa" (serratus, ventral, cervical and thoracic muscles) and "secreto" (*latissimus dorsi* muscle) from Iberian pig in order to test the effect of retail cut on the juiciness of the Iberian pork meat.

Finally, a preference analysis between sirloin (*iliopsoas* and *psoas minor* muscle) from Iberian pigs feeding with acorn and grass ("bellota") or with concentrates ("cebo") was performed by consumers.

4. Cooking and serving

Sensory analyses were carried out on samples after about 10 month frozen storage. Vacuum-packed samples were thawed for 24 to 36 h at 4ºC. After thawing, the muscles were cut in steaks (1 cm thick) and cooked to a final internal temperature of 72ºC using a two-sided electric grill (GV2PSG, Clajosa, Barcelona, Spain).

The samples were immediately served to consumers for their evaluation. For rinsing the mouth between samples, mineral water at room temperature and unsalted toasted bread were served to them.

III – Results and discussion

Characteristics of the consumer panel are summarized in Table 1. The tasters were mainly men over 30 years, regular consumers of pork meat.

Results of triangle tests indicated that there was a detectable difference ($p<0.01$) between the loin from Iberian pigs and heavy white pigs reared under similar conditions.

Table 1. Demographic characteristics and pork meat consumption frequency (n=85, %).

Age			Gender		Frequency of consumption			
20-30	**31-60**	**>60**	**Male**	**Female**	**>1/week**	**1/week**	**1-3/month**	**<1/month**
19.6	71.4	8.9	82.2	17.8	41.1	21.5	16.0	21.5

According to the tasters, texture characteristics were the main parameter that allowed differentiates the samples (Fig. 1). The tasters pointed out that the Iberian loin was more juicy and tender and showed higher flavour intensity than loin from white pig. These characteristics may be due to variations in intramuscular fat content of meat. Intramuscular fat influence on eating quality of meat resulted in higher saliva excretion during chewing which increase the juiciness of meat (Daszkiewicz *et al.,* 2003). Moreover, intramuscular fat facilitates separation of muscular filament and reduces the cutter force (Wood *et al.,* 1994). As has been pointed out by López-Bote (1998), Iberian breed pig has a high tendency to accumulate fat, whereas the other commercial breeds increases the lean content of the carcass and concomitantly decreases the intramuscular fat content (Ventanas *et al*., 2007).

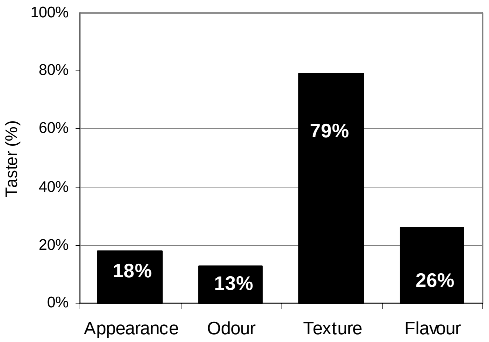

Fig. 1. Sensory differences indicated by the tasters (%) in the triangle test.

Concerning the results of the rank order test, differences ($p<0.05$) among the three retail cuts evaluated were detected. Results revealed that subjects considered "*pluma*" as the juiciest. However, there were not significant differences in the juiciness between "*presa*" and "*secreto*". A positive impression of juiciness of meat is related to high fat content (Jaworska et al., 2009). Results from the ranking test may be explained taking into account the inter and intramuscular fat content of the retails cuts evaluated. However, as far as we know, no papers have been devoted to the characterization of this kind of meat.

Regarding the preference tests, differences were not detected ($p>0.05$). The feeding of Iberian pig with acorn ("*bellota*") or concentrates ("*cebo*") did not suppose any preference of the consumers. This result would be due to that, today, Iberian pigs reared indoors are fed on monounsaturated fatty acid enriched concentrates (with high oleic acid sunflower oil) for obtaining a similar muscle fatty acid profile to that to pigs fed on acorns.

IV – Conclusions

On the basis of the results obtained it is possible to confirm that breed and muscle type affect sensory quality of pork meat after heat treatment. However, in this study the feeding on acorns did not contribute to increase consumers' preference of cooked Iberian meat.

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Influence of genetic type on the characteristics of subcutaneous adipose tissue of pig thighs destined for the PDO production

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Abstract. Italian heavy pig production is founded both on traditional breeds, such as Large White, Landrace and Duroc selected in Italy, and on hybrid pigs coming from specific plans of selection and crossbreeding. The aim of this work was to compare the characteristics of subcutaneous adipose tissue of fresh thighs obtained from pigs belonging to two genetic types and reared in the same farm. Thighs were destined to the production of PDO Italian dry-cured hams. Samples of subcutaneous adipose tissue from 46 thighs of traditional pigs (Italian Landrace x Large White cross) and from 32 thighs of Goland commercial hybrid line, were analysed for water and fat contents, fat iodine value and fatty acid composition. On the whole, hybrids showed, in comparison with traditional pigs, similar lipid content of subcutaneous adipose tissue but different lipid composition: higher levels of polyunsaturated fatty acids (141.1 *vs* 102.8 mg/g fat; P<0.001), lower levels of saturated (334.9 *vs* 371.0 mg/g fat; P<0.001) and mono-unsaturated (420.8 *vs* 452.5 mg/g fat; P<0.001) fatty acids, and greater iodine value (68.9 *vs* 62,4; P<0.001). Females showed lower fat content and higher degree of lipid unsaturation than castrated males.

Keywords. Italian heavy pig – Fresh ham – Lipid content – Fatty acid composition.

Influence du type génétique sur les caractéristiques du tissu adipeux sous-cutané des cuisses de porc destinées à la production sous AOP

Résumé. *La production italienne de porc lourd est fondée à la fois sur des races traditionnelles sélectionnées en Italie (Large White, Duroc et Landrace), et sur des sujets hybrides provenant de plans spécifiques de sélection et de croisement. Le but de ce travail était de comparer les caractéristiques du tissu adipeux sous-cutané des cuisses fraîches obtenues à partir de sujets appartenant à deux différents types génétiques élevés dans la même ferme. Les cuisses étaient destinées à la production de jambon sec italien AOP. Les échantillons de tissu adipeux sous-cutané de 46 cuisses de porc traditionnel (Landrace italien x Large White) et de 32 cuisses de porc hybride commercial Goland, ont été analysés pour la détermination des teneurs en eau et lipides, ainsi que la composition en acides gras et l'indice d'iode. Dans l'ensemble, les hybrides ont montré, en comparaison avec les porcs traditionnels, la même teneur en lipides mais des différences de composition en acides gras: le tissu adipeux sous-cutané des hybrides est plus riche en acides gras polyinsaturés (141,1* vs *102,8 mg/g lipides; P<0,001) et corrélativement plus pauvre en acides gras saturés (334,9* vs *371,0 mg/g lipides; P<0,001) et monoinsaturés (420,8* vs *452,5 mg/g lipides; P<0,001) et l'indice d'iode est plus élevé (68,9* vs *62,4 ; P<0,001). Les femelles ont montré la plus faible teneur en lipides et le plus haut degré d'insaturation des lipides par rapport aux mâles castrés.*

Mots-clés. *Porc lourd Italien – Jambon frais – Teneur en lipides – Composition en acides gras.*

I – Introduction

The quantity and quality of adipose tissue in the carcass is of utmost importance in the production of Italian heavy pigs, since fat is a basic component of raw cuts destined to processing into valuable products, such as PDO dry cured hams (Lo Fiego *et al.*, 2005; Lo Fiego *et al.*, 2010). It is well known that lipid composition of fat tissues of pigs is directly influenced by the fat composition of diets (Bee *et al*., 2002, Rossi and Corino, 2002; Kouba *et al*., 2003), by genetic type (Bout *et al*., 1988; Lo Fiego *et al*., 2005) and by carcass fatness (Scott *et al*., 1981; Lo Fiego, 1996; Lo Fiego *et al.*, 2010). In the past years Italian heavy pig production has undergone significant changes due to the massive introduction of more recent genetic types, for the largest part commercial hybrids. Moreover, the “traditional” breeds, thanks to constant and continuous selection programs, optimized diets and improved rearing techniques, have themselves been modified, and carcass fat content has been reduced. Both traditional breeds, such as Large White, Landrace and Duroc selected in Italy, and hybrid pigs, coming from specific plans of selection and crossbreeding, have been used since many years for typical productions. Hybrids offer appreciable performance traits, but often show different composition of fat depots in comparison with traditional types: in particular, their growing use in heavy pig production produced an increase of the degree of lipid unsaturation, which is not desired for typical productions. Due to the necessity of constantly monitoring the quality of raw materials obtained from animals which are subject to continuous evolution, the aim of this work was to compare the characteristics of subcutaneous adipose tissue of fresh thighs destined to the production of PDO Italian dry-cured hams, and obtained from pigs belonging to two genetic types and reared in the same condition in the same farm.

II – Material and methods

For this research, a total of 78 left thighs destined to the PDO Italian ham dry-curing production were used. Samples of subcutaneous adipose tissue from 46 thighs of traditional pigs (Italian Landrace x Large White cross, 34 castrated males and 12 females) and from 32 thighs of Goland commercial hybrid line (22 castrated males and 10 females), reared in the same farm and fed with a standard cereals-soybean meal based commercial feed, were analysed for water content, fat content and fat iodine value as reported by Lo Fiego *et al*. (2005). Moreover, fatty acid composition of lipid was determined using a TRACE™ GC Ultra (Thermo Electron Corporation, Rodano, Milano, Italy) equipped with the Ultra Fast Module (UFM), a Fast Flame Ionization Detector and a UFM-Carbovax column, 5 m long, 0.1 mm i.d, 0.2 µm film thickness as described in a previous paper (Ficarra *et al*., 2010). The FAME were identified by comparison of each retention time with the known retention times of the corresponding pure standards (Supelco 37 Component FAME mix and PUFA standard n. 2, animal source, Supelco, Bellafonte, PA, USA). For quantification purposes, the response factor was calculated and the method of the internal standard was used. The results were expressed as mg of each fatty acid methyl ester/g of lipids. Data were subjected to analysis of variance using the GLM procedures of the SAS statistical package (SAS, 1996) and a factorial model that considered genetic type and sex as fixed effects.

III – Results and discussion

Table 1 shows carcass and raw ham weights and fat thickness measurements according to genetic type. The mean values shown in Table 1 for both genetic types represent the optimum for Italian heavy pig production. No statistical difference was found between genetic types as regard both carcass and raw ham refrigerated weight and carcass backfat thickness. The latter was, however, slightly higher in traditional pigs, whose subcutaneous ham backfat thickness was lower in comparison with hybrids (- 4 mm, $P<0.01$), with subsequent lower trimming loss ($P<0.01$). This shows how the dislocation of fat depots in the carcass may vary in different genetic types, and consequently carcass backfat thickness might be not suitable for the estimation of fatness in other cuts. About the effect of sex (data not reported in the tables), females showed lower carcass weight (-10.4 kg; $P<0.01$), lower refrigerated (-0.9 kg; $P<0.01$) and trimmed ham weight (-0.7 kg; $P<0.05$), and thinner subcutaneous fat (- 4.3 mm; $P<0.05$) in comparison with castrated males. Table 2 shows the water and ether extract content and fatty acid composition of the subcutaneous adipose tissue of raw ham according to genetic type. Genetic type did not affect water and ether extract: on the opposite, in a previous work, hybrids, belonging to a different genetic type, showed higher water and lower lipid content in comparison with traditional pigs reared in the same condition (Lo Fiego *et al.*, 2005). The fatty acid composition of the lipids was significantly influenced by genetic type.

Table 1. Effect of genetic type on carcass and raw ham traits

	Genetic type		
	Traditional	Commercial Hybrid	R -MSE
Refrigerated carcass weight (kg)	127.8	128.6	10.96
Carcass backfat thickness† (mm)	25.9	24.0	4.83
Refrigerated ham weight (kg)	16.8	16.4	1.34
Trimmed ham weight (kg)	13.9^{a}	13.2^{b}	1.09
Subcutaneous ham fat thickness†† (mm)	33.9^{A}	37.9^{B}	7.23
Trimming loss (%)	17.9^{A}	18.5^{B}	2.47

†Measured between the ¾ last rib at 8 cm from the splitting line of the carcass.
††Measured beneath the femur head.
$^{A, B}$ = P≤0.01, $^{a, b}$ = P≤0.05.

Table 2. Effect of genetic type on water and ether extract contents (%), lipid fatty acid composition (mg/g of lipids) and iodine value of covering adipose tissue of raw ham

	Genetic type		
	Traditional	Commercial Hybrid	R -MSE
Water content %	6.57	6.96	1.64
Ether extract content %	90.11	90.91	3.12
Fatty acids composition† mg/g			
C14:0	12.55	11.93	1.68
C16:0	220.78^{A}	209.48^{B}	15.14
C18:0	131.40^{A}	108.13^{B}	11.28
C20:0	1.95^{A}	1.59^{B}	0.34
Total saturated	371.04^{A}	334.93^{B}	22.10
C16:1	21.21	22.96	3.58
C18:1	417.96^{A}	386.10^{B}	22.30
C20:1	9.13^{A}	7.69^{B}	1.51
Total monounsaturated	452.53^{A}	420.77^{B}	24.34
C18:2	91.81^{A}	125.33^{B}	14.60
C18:3	4.27^{A}	5.56^{B}	0.78
C20:2	4.50^{A}	5.65^{B}	0.80
C20:4	1.43	1.59	0.40
Total polyunsaturated	102.84^{A}	141.12^{B}	15.63
Iodine value	62.39^{A}	68.91^{B}	2.95
Total fatty acids %			
Saturated	40.18^{A}	37.46^{B}	1.82
Monounsaturated	48.66^{A}	46.80^{B}	1.92
Polyunsaturated	11.16^{A}	15.74^{B}	1.55

$^{A, B}$ = P≤0.01; $^{a, b}$ = P≤0.05.
†Other fatty acids detected: $C_{17:0}$, $C_{17:1}$, $C_{18:3\_n6}$, $C_{20:3}$.

Traditional pigs produced lipids with a higher content of palmitic ($C_{16:0}$), stearic ($C_{18:0}$) and eicosanoic ($C_{20:0}$) fatty acids (P<0.01) and showed, consequently, a higher level of total saturated fatty acids (SFA) (452.5 *vs* 420.8 mg/g of lipids; P< 0.01). Total monounsaturated fatty acids (MUFA) were also significantly higher in these subjects (P< 0.01) and the differences were mainly due to oleic ($C_{18:1}$) and eicosenoic fatty acids ($C_{20:1}$). The hybrid pigs were found to have a significantly higher polyunsaturated fatty acid content (PUFA) (141.1 *vs* 102.8 mg/g of lipids; P< 0.01), mainly linoleic ($C_{18:2}$) (125,3 *vs* 91,8 mg/g of lipids), linolenic ($C_{18:3}$) (5,6 *vs* 4,3 mg/g of lipids) and eicosadienoic ($C_{20:2}$) (5,7 *vs* 4,5 mg/g of lipids). Consequently, the hybrids also showed a higher iodine value (68.9 *vs* 62.4; P< 0.01). Linoleic and stearic fatty acids are the main components which may influence technological characteristics of lipids, such as firmness and cohesiveness; moreover an excessive content of $C_{18:2}$ may favor oxidative phenomena during seasoning period. Based on the mean of $C_{18:0}$ and $C_{18:2}$ content and iodine value, it can be stated that both genetic types fall within the limits set by Italian PDO ham Consortia (Lo Fiego *et al.*, 2005). As regard the sex (data not reported in the table), females showed lower ether extract content and higher content of total polyunsaturated fatty acids, mainly $C_{18:2}$, $C_{18:3}$ and $C_{20:2}$, than castrated males (P< 0.01).

IV – Conclusions

The results show that the fatty acid composition of subcutaneous adipose tissue of fresh ham from heavy pigs may be considerably influenced by genetic type. The hybrid pigs, confirming the results of previous research on heavy pigs, were found to have a significantly higher polyunsaturated fatty acids content respect to the traditional pigs coming from the national selection programs; but the subjects examined in this research are anyway suitable for PDO production. Thus, taking into account the excellent on-farm performance of hybrids, it seems necessary a strict control of fat depots traits, in order to avoid an excessive increase of unsaturation degree.

Acknowledgements

The Authors want to thank AGRIRICO S.R.L. (Formigine – Modena) and ITALCARNI S.C.A. (Carpi-Modena) for pig supply and slaughtering. The research was supported by MIUR- PRIN 2007 (Prof. D.P. Lo Fiego).

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Effets de la supplémentation alimentaire en acide pantothénique sur la composition en acides gras de la bardière chez le porc lourd en finition

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Resumé. Cette étude a évalué l'effet de la supplémentation alimentaire en acide pantothénique sur la composition en acides gras (AG) de la bardière chez le porc lourd en finition. Dans ce but, 42 porcs, 21 femelles et 21 mâles castrés de poids vif et d'âge similaires, ont été uniformément allotés en trois groupes de 14 sujets chacun. A partir de 95 kg de poids vif (PV) jusqu'à l'abattage (165 kg PV), les 3 groupes ont reçu le même régime de base mais contenant différents niveaux d'acide pantothénique, sous forme de calcium pantothénate: respectivement 10 (contrôle, C), 60 (T1) et 110 (T2) ppm. Lors de l'abattage, des échantillons de gras de la bardière ont été prélevés individuellement, puis soumis à l'analyse de la composition en AG, par chromatographie capillaire en phase gazeuse, et à la détermination de l'indice d'iode selon la méthode de Wijs. L'utilisation de doses élevées d'acide pantothénique dans la ration conduit à une diminution des AG saturés (P=0,061) et monoinsaturés (P=0,098) de la bardière, tandis que les AG polyinsaturés et l'indice d'iode augmentent (P<0,05). L'acide pantothénique administré à des concentrations supérieures à 60 ppm n'a pas entraîné d'effets significatifs sur la composition en AG de la bardière. Globalement, la qualité technologique du gras de la bardière chez le porc lourd pourrait être négativement affectée par l'utilisation alimentaire de niveaux élevés d'acide pantothénique.

Mots-clés. Porc lourd – Ration de finition – Acide pantothénique – Bardière – Acides gras.

Effects of dietary pantothenic acid supplementation on fatty acid composition of backfat in the finishing heavy pig

Abstract. *This study evaluated the effect of pantothenic acid supplementation to the finishing diet of heavy pigs on backfat fatty acid (FA) composition. To this aim, 42 pigs, 21 females and 21 castrated males of similar weight and age, were evenly divided into three groups of 14 subjects each. From 95 kg live weight (lw) till slaughtering (165 kg lw), the 3 groups received the same basal diet containing either 10 (control, C), or 60 (T1) or 110 ppm (T2) of pantothenic acid, as calcium pantothenate. At slaughter, backfat samples were individually collected and then submitted to FA composition analyses, by capillary GLC. The feeding of vitamin B5 at levels higher than currently recommended led to a decrease of saturated (P=0.061) and monounsaturated FA (P=0.098) content in backfat lipids, whereas polyunsaturated FA content and the iodine value increased (P<0.05). Besides, the feeding of pantothenic acid at levels higher than 60 ppm brought about no further relevant effect on FA content. On the whole, the technological quality of backfat lipids in the heavy pig could be negatively, albeit slightly, affected by high level of pantothenic acid in the finishing diet.*

Keywords. *Heavy pig – Finishing diet – Pantothenic acid – Backfat lipids – Fatty acids.*

I – Introduction

Des études récentes ont montré que l'utilise de doses élevées d'acide pantothénique (vitamine B5) dans l'aliment entraîne un effet favorable sur la vitesse de croissance et sur la valeur commerciale de la carcasse avec une augmentation de la proportion des tissus maigres et une réduction de la masse adipeuse (Autrey *et al.*, 2002; Santoro *et al.*, 2006, Lo Fiego *et al*., 2009). Toutefois cette diminution pourrait comporter une augmentation du degré d'insaturation du tissu adipeux (Lebret et Mourot, 1998) avec conséquences défavorables sur sa qualité technologique (Lo Fiego *et al*., 2005). A ce jour, l'effet de la supplémentation alimentaire de la vitamine B5 sur les caractéristiques des tissus adipeux du porc lourd n'a pas été étudié.

L'objectif de cette recherche était donc d'évaluer l'influence des niveaux élevés d'acide pantothénique, dans la ration du porc lourd en finition, sur le profil d'acides gras de la bardière.

II – Matériels and méthodes

L'essai était conduit sur 42 animaux (Dumeco-Cofok x (LxLW)), dont 21 femelles et 21 mâles castrés, uniformément allotés, par poids et par sexe, en trois groupes de 14 porcs chacun. À partir de 95 kg de poids vif les animaux ont été alimentés avec la même ration composée de céréales, tourteau de soja et mélasse (MAT 15%), mais additionnée avec différents apports de pantothénate de calcium, pour obtenir une dose d'acide pantothénique de 10 (contrôle, C), 60 (T1) et 110 (T2) ppm. Les animaux ont été abattus lorsqu'ils ont atteint un poids vif d'environ 165 kg. Des prélèvements de la bardière ont été effectués dès l'abattage. Les échantillons ont été stockés à -20°C dans l'attente des analyses. Les lipides totaux ont était extraits selon la méthode IUPAC (1979). La composition en acides gras des lipides de la bardière a était déterminée par chromatographie capillaire en phase gazeuse. L'indice d'iode a été déterminé selon la méthode de Wijs. Les résultats ont était analysés à l'aide de l'ANOVA en considérant le niveau alimentaire d'acide pantothénique comme une variable indépendante (SAS, 1996).

III – Résultats et discussion

De façon générale l'accroissement de l'acide pantothénique dans la ration du porc lourd en finition s'accompagne d'une diminution de l'épaisseur de la bardière (P=0,09) et de l'incidence de morceaux adipeux (P<0,05) de la carcasse (Tableau 1). Ainsi, les données obtenues confirment la capacité de la vitamine B5 à réorienter l'énergie alimentaire au détriment du tissu adipeux. L'utilise des doses élevées d'acide pantothénique dans la ration a influencé la composition en acides gras de la bardière (Tableau 2). Globalement on a constaté une réduction des acides gras saturés (P=0,061), monoinsaturés (P=0,098) et une augmentation des polyinsaturés totaux et de l'indice d'iode (P<0,05), indépendamment du dosage.

Tableau 1. Caractéristiques de la carcasse

	Intégration alimentaire en acide pantothénique (ppm)					
	10	60	110	C *vs* moyenne T1+T2	T1 *vs* T2	R-MSE
	C(n=14)	T1(n=14)	T2(n=14)			
Poids de la carcasse (kg)	142,5	138,7	140,4	ns	ns	8,66
Viande maigre (%)	47,9	49,5	48,8	ns	ns	3,05
Epaisseur moyenne de la bardiére (mm)	31,5	27,0	28,2	$	ns	6,26
Morceaux maigres† (%)	64,8	65,7	65,8	ns	ns	2,46
Morceaux gras† (%)	33,3	31,5	30,6	*	ns	3,04

†% de poids de carcasse chaude;
*= P<0,05; $=P<0,1; ns = pas significatif.

Plus en détail, l'ajoute de doses élevées d'acide pantothénique a déterminé une diminution de la teneur en acide palmitique (P<0,01), bien connu pour ses propriétés athérogènes. Par contre le niveau d'acide stéarique n'est pas influencé par la teneur en vitamine B5 de la ration. Il montre des valeurs supérieures à 12% des acides gras totaux, c'est-à-dire au dessus de la teneur minimale recommandée pour satisfaire les caractéristiques technologiques et organoleptiques du tissu adipeux (Lebret et Mourot, 1998).

Tableau 2. Effet de la teneur en acide pantothénique sur la composition en acides gras de la bardière (mg/g des lipides)

	Intégration alimentaire en acide pantothénique (ppm)					
	10	60	110	C vs moyenne T1+T2	T1 *vs* T2	R-MSE
	C(n=14)	T1(n=14)	T2(n=14)			
Acides gras (mg/g des lipides)						
C14:0	11,26	10,63	10,71	*	ns	0,86
C16:0	222,93	213,56	213,85	**	ns	9,72
C17:0	2,66	2,72	2,52	ns	ns	0,43
C18:0	131,92	131,09	131,23	ns	ns	8,25
C20:0	1,65	1,73	1,86	ns	ns	0,50
Total saturés	370,42	359,73	360,17	$	ns	15,17
C16:1	17,85	16,48	15,92	**	ns	1,80
C17:1	2,25	2,18	1,85	ns	$	0,43
C18:1	394,18	384,58	390,82	ns	ns	15,87
C20:1	10,92	8,45	10,54	**	**	1,51
Total monoinsaturés	425,20	411,69	419,13	$	ns	17,03
C18:2	169,86	183,79	180,33	*	ns	14,99
C18:3	8,93	10,36	9,09	$	*	1,18
C20:2	7,51	8,41	8,20	*	ns	0,94
C20:3	1,25	1,75	1,61	ns	ns	0,71
C20:4	1,75	2,06	1,82	ns	ns	0,66
Total polyinsaturés	189,30	206,37	201,05	*	ns	16,94
Indice d'iode	70,41	72,26	71,91	*	ns	2,55
Total acides gras %						
Saturés	37,61	36,79	36,74	$	ns	1,43
Monoinsaturés	43,17	42,10	42,75	ns	ns	1,43
Polyinsaturés	19,22	21,11	20,51	**	ns	1,65

**= P<0,01; *= P<0,05; $=P<0,1; ns = pas significatif.

Parmi les acides gras polyinsaturés, l'acide linoléique a montré l'augmentation la plus importante (P<0,05) à des doses élevées d'acide pantothénique avec des valeurs au dessus de 15%, teneur maximale recommandée pour éviter les problèmes lors de la transformation et de la conservation des produits (Wood, 1984). Mais cette valeur était déjà plus élevée même dans le groupe de contrôle.

IV – Conclusions

Les résultats obtenus avec cette première étude sur l'influence de la supplémentation alimentaire de l'acide pantothénique sur les caractéristiques des tissus adipeux du porc lourd ont mis en évidence que l'addition de doses élevées d'acide pantothénique implique une réduction de l'incidence de morceaux gras et de l'épaisseur de la bardière avec une augmentation du degré d'insaturation des lipides. Dépassant les 60 ppm n'entraîne pas d'autres changements. Cela répond mieux aux besoins spécifiques des consommateurs qui donnent plus d'importance à la composante nutritionnelle des aliments, mais au contraire n'est pas accueil pour les transformateurs qui sont plus exigeants sur l'aptitude à la transformation et à la conservation des tissus adipeux.

Remerciements

Les auteurs remercient Dr. A. Filippini, de prendre soin des animaux, et Raggio di Sole S.p.A., pour formuler et fournir les aliments.

Ce travail a reçu le soutien financier de la Fondation Pietro Manodori, Reggio Emilia et RFO, Università di Bologna.

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Physical measures of the carcass and the chemical composition of *Longissimus dorsi* muscle of Alentejano pigs between 70 and 110 kg LW

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Abstract. The aim of this work was to determine the relationship between physical measures from the subcutaneous tissue and Longissimus dorsi (LD) muscle (area, depth, and width measured between the 3rd and 4th lumbar vertebrae, at the last rib, and between the 3rd and 4th last ribs) and the chemical composition of LD at 70, 80, 90, 100, and 110 kg LW. The content of water, protein, neutral and polar lipids, total and soluble collagen, and total pigments, were determined. Globally, the measures taken and the chemical composition were not affected between 70 and 110 kg, except for the LD depth and width at the 3rd-4th lumbar vertebrae. At 70 kg, the LD depth was greater than at 110 kg (3.77 vs 2.75 cm, $P<0.05$) and the width was smaller (8.14 vs 9.82 cm, $P<0.05$). In conclusion, from 70 to 110 kg: (i) the morphological changes in the lumbar region were due mainly to the width dimension, with no impact on the chemical composition of the muscle; and (ii) the chemical composition did not change drastically, even though the amount of intramuscular fat increased slightly between 70 and 110 kg (5.32 and 6.67%, respectively) suggesting an early intramuscular fat deposition.

Keywords. Alentejano pig – Carcass – Meat quality – Live weight.

Mesures physiques de la carcasse et composition chimique du muscle* Longissimus dorsi *des porcs Alentejano entre 70 et 110 kg de poids vif

Résumé. *Le but de ce travail était de déterminer la relation entre les mesures physiques du tissu sous-cutané et le muscle* Longissimus dorsi *(LD) (superficie, profondeur et largeur mesurées entre les 3e et 4e vertèbres lombaires, à la dernière côte, et entre les dernières 3e et 4e côtes) et la composition chimique du LD à 70, 80, 90, 100, et 110 kg de poids vif. Les teneurs en eau, protéines, lipides neutres et polaires, collagène total et soluble, et pigments totaux ont été déterminées. Globalement, les mesures prises et la composition chimique n'ont pas été modifiées entre 70 et 110 kg, sauf pour la profondeur et la largeur du LD au niveau des vertèbres lombaires 3e-4e. À 70 kg, la profondeur du LD a été supérieure (3,77* vs *2,75 cm, P <0,05) et la largeur inférieure (8,14* vs *9,82 cm, P <0,05) par rapport à 110 kg. En conclusion, entre 70 et 110 kg: (i) les changements morphologiques dans la région lombaire ont été principalement attribuables à la dimension de la largeur, sans impact sur la composition chimique du muscle; et (i) la composition chimique ne change pas radicalement, même si la quantité de lipides intramusculaires a légèrement augmenté entre 70 et 100/110 kg (5,32 et 6,42/6,30%, respectivement) suggérant un dépôt de gras intramusculaire précoce.*

Mots-clés. *Carcasse – Composition chimique – Alentejano.*

I – Introduction

For centuries, the agriculture in Alentejo region was based in a production system characterised by a strong interdependence between natural fed resources and animal production. In this context, the Alentejano pig breed gained importance by its rusticity and the rational use of fed resources. Comparatively with others, the Alentejano pig shows a slow rate of growth and a high lipogenesis activity at early stages of development. The lipids are deposited mainly in the subcutaneous, renal and pelvic regions. The percentages of fatty cuts can attain more than 50%

of the carcass weight and the backfat thickness at the last rib can reach 60 mm at 120 kg live weight (LW) (Almeida *et al*., 1993; Neves *et al*., 2001). Nowadays, the production fulfills a double function: it provides meat for the manufacture of cured products and also for fresh consumption. Considering the volume and the increase of fresh pig meat consumption in Portugal, the farmers have increased their production for this market, traditionally dominated by the meat from precocious pig breeds reared in intensive production systems. Based on high organoleptic quality, pork from Alentejano hogs could be an effective alternative for meat from industrial breeds. However, carcass has too much fat and a bad proportion of lean cuts, which impair the economic rentability of this production system. The animal growth implies chemical, biochemical, and physical changes in the adipose and muscular tissues, mainly due to an increase on the lipid content (subcutaneous and intramuscular) (Mayoral *et al*., 1999). These changes affect the gross chemical composition. Thus, it was interesting to find if the growth process and fat deposition induce significant changes on muscle chemical composition that could influence its global quality. Since it is possible to measure the depth of LD *in vivo*, it was interesting to study the possible relationship between this measure and the chemical composition of the muscle, particularly with the amount of intramuscular fat at the usual commercial slaughter weights.

II – Materials and methods

Thirty Alentejano pigs were castrated at the age of 60 days. After weaning, piglets were transferred to individual pens at open air and fed a commercial diet (15% CP; 3100 kcal DE) offered at 85 % of *ad libitum*. At the beginning of the experiment, 5 animals with 40 kg LW were slaughtered. The remaining animals were weighted weekly and slaughtered (5 animals) at 70, 80, 90, 100, and 110 kg LW. After 12 h fasting, the animals were weighted and then slaughtered. Twenty four hours after chill, the left side of each carcass was submitted to commercial cuts, according to the Portuguese Norm N – 2931. The measure of loin muscle depth was done 6 cm lateral to the mid-line of the carcass and resulted from the average of the first measure and two others 0,5 cm apart from it in both directions. The loin muscle area and width were also measured. Those measures were taken at three anatomical sites (between the 3rd and 4th lumbar vertebrae, the last rib and between the 3rd and 4th last ribs). Samples were taken from the loin cut and were vacuum packaged and stored (-30ºC) until analysis. Moisture (Portuguese Norm 1614), total protein (Portuguese Norm 1612), total, neutral, and polar lipids (Marmer and Maxwell, 1981), and total pigment (Hornsey, 1956) were determined. Total hydroxyprolin was analyzed (Woessner, 1961) and multiplied by 7.14 (Etherington & Sims, 1981) to obtain the total collagen. The soluble fraction of collagen was also analyzed (Hill, 1966). An ANOVA was carried out and the means comparison was made by SNK test. The correlation between the variables studied was determined by the Pearson coefficient. SPSS statistical program was used.

III – Results

Globally, the chemical composition and the measures taken weren't affected by live weight between 70 and 110 kg (Tables 1 and 2), except for the depth and width of LD between the 3rd and 4th lumbar spot (Table 2). At 70 kg the depth of the LD was greater ($P<0.05$) than at 110 kg LW (3.77 cm *vs* 2.75 cm) and the width was smaller (8.14 cm *vs* 9.82 cm). In the other anatomical sites, no significant differences were found. However, the LD area at the last rib tended to be greater ($P=0.052$) at 110 kg than at 70, 80, and 90 kg LW (21.45 cm^2 *vs* 17.27, 17.68, and 17.56 cm^2, respectivelly). The average area, depth and width presented no significant differences between 70 and 110 kg LW (17.77 cm^2 *vs* 20.76 cm^2 for area; 3.52 cm *vs* 3.32 cm for depth, and 7.42 *vs* 8.37 cm for width). However, the P value obtained for the width was 0.057 which seems to indicate that the increase in the muscle area was due essentially to the increase in width dimension. Daza *et al*., (2006) observed in Iberian pigs reared extensively and slaughtered at 152 kg LW, a LD area measured at last rib level of 26.73 cm^2 , and in pigs in confinement slaughtered at 159 kg, 30.31 cm^2. These values, greater than those observed on this work, could be justified by the greater slaughter weight. Nevertheless, they showed a development pattern similar to the one observed in the present work.

Table 1. Chemical characteristics of *Longissimus dorsi* muscle of Alentejano pigs at various live weights

	Slaughter weight groups (kg LW)				
	70	**80**	**90**	**100**	**110**
Water	69.97 ± 1.7	70.33 ± 0.3	67.39 ± 2.1	67.24 ± 2.5	68.70 ± 0.8
Protein	23.07 ± 0.6	22.41 ± 1.4	22.50 ± 0.9	22.97 ± 0.6	21.86 ± 0.7
Neutral lipids	5.37 ± 2.3	5.23 ± 0.5	6.24 ± 1.3	6.42 ± 1.2	6.67 ± 1.5
Polar lipids	0.85 ± 0.1^{ab}	1.18 ± 0.1 ^{ab}	0.99 ± 0.2^{ab}	0.73 ± 0.2^{b}	0.86 ± 0.1^{ab}
Total pigment	69.53 ± 2.8	67.32 ± 5.9	84.18 ± 11.7	71.90 ± 17.1	73.78 ± 10.3
Total collagen (ug/mg, DM)	12.47 ± 4.0	9.99 ± 1.0	10.84 ± 1.0	8.98 ± 2.0	10.23 ±.2.0
Soluble collagen (ug/mg, DM)	1.93 ± 1.0	1.20 ± 0.1	0.76 ± 0.3	1.17 ± 0.2	0.97 ± 0.4

Means in the same line with different letter are significantly different (P<0.05).

Table 2. Physical characteristics of the backfat of Alentejano pigs at a various live weights

	Slaughter weight groups (kg LW)				
	70	**80**	**90**	**100**	**110**
3th and 4th LuVA (cm^2)	18.70 ± 1.9	19.45 ± 2.3	21.32 ± 2.1	20.66 ± 1.7	20.94 ± 4.1
3th and 4th LuVD (cm)	3.77 ± 0.7 ^{b}	2.99 ± 0.1 ^{b}	3.12 ± 0.2 ^{b}	3.00 ± 0.1 ^{b}	2.75 ± 0.3 ^{a}
3th and 4th LuVW (cm)	8.14 ± 0.4 ^{a}	8.73 ± 0.6 ^{a}	8.41 ± 1.1 ^{a}	8.54 ± 0.3 ^{a}	9.82 ± 1.1 ^{b}
LaRA (cm^2)	17.27 ± 2.4	17.68 ± 2.8	17.56 ± 1.4	19.42 ± 2.8	21.45 ± 1.7
LaRD (cm)	3.41 ± 0.2	3.43 ± 0.5	3.30 ± 0.5	3.17 ± 0. 5	3.63 ± 0.5
LaRW (cm)	7.28 ± 0.5	7.46 ± 0.3	7.08 ± 0.6	7.64 ± 0.5	7.82 ± 0.5
3th and 4th LaRA (cm^2)	17.35 ± 2.9	18.57 ± 2.9	20.49 ± 2.5	19.35 ± 1.9	19.88 ± 1.4
3th and 4th LaRD (cm)	3.38 ± 0.2	3.57 ± 0.2	3.46 ± 0.4	3.53 ± 0.4	3.59 ± 0.3
3th and 4th LaRW(cm)	6.84 ± 0.2	7.37 ± 0.4	7.37 ± 0.9	6.68 ± 0.2	7.48 ± 0.6

Means in the same line with different letter are significantly different (P<0.05).
LuVA: lumbar vertebrae area; LuVD: lumbar vertebrae depth; LuVW: lumbar vertebrae width; LaRA last ribs area; LaRD: last ribs depth; LaRW: last ribs width.

The amount of neutral lipids showed a ~20% increase between 70 and 100/110 kg LW(5.32% vs 6.42/6.30 %, respectively). The same was observed in a previous work by Neves *et al*., (1996). However, Estevez *et al*. (2003) reported in three genetic lines intramuscular fat contents between 2.51 and 3.34% in pigs with 90 kg LW. Those amounts, substantially lower than the ones obtained in this work, could be explained by some genetic selection in the lines of the Iberian pigs studied in that work. The correlation analysis showed a negative relation between neutral intramuscular fat and protein (-0.557; P<0.01) of the LD muscle, as expected. The average area showed a greater correlation coefficient with the area of LD measured at the last rib (0.873), than with the areas measured at the two other spots (0.821 and 0.798 for the 3rd and 4th lumbar vertebrae spot and the 3rd and 4th last ribs spot, respectively). The correlation study between the average area and the individual measure of depth and width in the three spots shown only significant correlation with the depth of LD at the last rib (0.399; 0.048) and with the width measured between the 3rd and the 4th lumbar vertebrae (0.464; 0.020). Finally, no correlations were observed between the chemical composition traits and the physical measures taken in the LD muscle.

IV –Conclusions

Between 70 and 110 kg LW, (i) the lumbar region was the one that showed more morphological changes during growth, and the development of the muscle seems to be due to width more than the depth dimension; (ii) the chemical composition didn't change much, and in the particular case of the amount of intramuscular fat, only a slight increase between 70 and the heavier weights (100 and 110 kg LW) was observed, suggesting a earlier intramuscular fat deposition; and (iii) no statistical significant correlation was found between the chemical composition traits and the physical measures taken from the backfat and from the LD muscle.

Acknowledgements

The authors wish to acknowledge the financial support received from the Program Agro (Project 226).

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Consumers perception of iberian cooked meat products

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Abstract. Today, many consumers demand and have available a wide assortment of high quality foods. The quality of meat products is influenced by intrinsic factors such as breed, genotype, reared system, and feeding regime. Thus, cooked products manufactured from raw material from Iberian and white pigs should show different sensory quality. The aim of this study was to evaluate different cooked products made from Iberian and white pigs, in order to establish differences. Samples of different cooked meat products were purchased from local supermarkets (cooked ham, pâté and cooked sausages). In order to know if consumers could distinguish between Iberian and white cooked meat products, different tests (triangle test, duo-trio test and ranking test) were carried out with consumers. The results showed that consumers were able to distinguish between cooked meat products manufactured from Iberian and white raw meat. According to consumers, flavour and juiciness were the attributes that allowed distinguish these products.

Keywords. Sensory analysis – Cooked meat product – Iberian and white pig.

Perception qu'ont les consommateurs des produits cuits à base de viande de porc Ibérique

Résumé. *Les attributs qui sont propres aux produits de la viande et qui les rendent très appréciés par les consommateurs, sont étroitement liés aux caractéristiques des matières premières, notamment la génétique et le système d'exploitation des animaux. Pour cette raison, les produits cuits élaborés avec des matières premières de porc Ibérique devraient présenter des différences par rapport à ceux élaborés avec des matières premières de porc blanc. L'objectif de l'étude a été l'évaluation sensorielle de différents produits cuits élaborés avec du porc Ibérique ou du porc blanc, afin de pouvoir établir des différences. Les résultats obtenus ont montré que les consommateurs ont été capables de distinguer entre les produits élaborés avec une matière première de porc Ibérique et ceux à base de porc blanc. Les attributs qui ont permis d'établir des différences ont été le goût et la teneur en jus.*

Mots-clés. *Analyse sensorielle – Viande de porc Ibérique et de porc blanc – Produits cuits.*

I – Introduction

Cooked pork meats like pâté, sausages or cooked ham are very consumed in Spain. Even though there is a large variety of cooked products on the Spanish market, most of them are made using meat and back fat from white pigs. From a nutritional point of view, fat from white pigs has a considerable level of saturated fatty acids and cholesterol, with an undesirable n-6/n-3 polyunsaturated fatty acids ratio. Consumer concerns about the relationship between health and nutrition, challenge food technologists to develop new meat and fat-based products with improved characteristics.

In this sense, using meat and adipose tissue from Iberian pigs for the manufacture of cooked products results in a high quality product. Intramuscular fat of these pigs is characterized by large percentages of monounsaturated fatty acids, a small proportion of hypercholesterolemic fatty acids and present lower values of the ratio n-6/n-3 than fat from white pigs (Estevez *et al.,* 2006). The products traditionally obtained from these animals such as dry cured hams and dry-cured loins are highly appreciated by Spanish consumers. However, to date, the iberian cooked meat products are not well known by consumers.

The aim of this study was to evaluate if the consumers are able to establish differences between diverse cooked products made from Iberian and white pigs, in order to increase the knowledge about the consumers' perception to these new products.

II – Material and methods

To achieve this objective pâté, cooked ham and sausages manufactured from white or Iberian pigs were used. All samples were purchased from local markets and were subject to consumer sensory evaluation.

The tasters (n=84) were mainly farmers, manufacturers and technicians associated with pig meat industries, habitual consumers of meat products. Characteristics of the untrained panel are summarized in Table 1.

Table 1. Characteristics of the tasters based on age and gender

	Age (years)				
Tasters	**20-30**	**31-40**	**41-50**	**51-60**	**>60**
N†	21.4%	27.4%	29.8%	13.1%	8.3%
Male	11.9%	17.9%	22.6%	11.9%	8.3%
Female	9.5%	9.5%	7.2%	1.2%	–

†All consumers.

In order to know if consumers could establish differences between cooked meat products manufactured from white or Iberian pork meat, consumers carried out different discrimination tests. First, a triangle test was carried out according to the ISO 4120:2004. Samples, consisting of Iberian pâté and pâté were served on plastic plates to participants. Three coded samples were presented to each panellist and each panellist was asked to pick out which sample they feel different from the other two. Besides, to complement the triangle test, tasters were asked to indicate the reason for selecting one particular sample of the three used in the analysis. Then, consumers carried out a duo- trio test (ISO 10399:2004). Three samples of cooked ham (manufactured from white or Iberian pork meat) were presented to tasters: one sample was labeled "R" (reference) and the other two were coded. One of the coded samples was identical to "R" and panellists were asked to identify the correct sample. Besides, tasters were asked to indicate the reason for selecting one particular sample of the three used in the analysis. Finally, a ranking test (ISO 8587:2006) was carried out with three cooked sausages kind: Frankfurt, Bratwurst and iberian sausages. Panellists received three coded samples and were requested to rank samples for intensity of some specific characteristic. Besides, the subjects were asked to specify the characteristic that determined their preference.

Mineral water at room temperature and unsalted toasted bread were available and tasters were required to consume them before tasting each sample in order to rinse their mouths between samples. Consumer's tests were conducted in a room with white fluorescent lighting and kept at constant temperature of 21±2ºC at Salamanca (Spain).

III – Results and discussion

The results of the triangle test are shown in Table 2 and revealed that tasters perfectly discriminated among the tasted samples.

The age and the gender of consumers did not establish differences in the significance level. In general, all tasters chosen the flavour, principally, for selecting the iberian pâté as different sample (Fig. 1).

Table 2. Results obtained in the triangle test carried out with pâté

Age	Tasters	No. judgements corrects	Significance level
20-30	N†	18/18	***
	Male	10/10	***
	Female	8/8	***
31-40	N	21/23	***
	Male	13/15	***
	Female	8/8	***
41-50	N	21/25	***
	Male	16/19	***
	Female	5/6	**
51-60	N	11/11	***
	Male	10/10	***
	Female	1/1	—
>60	N	7/7	***
	Male	7/7	***
	Female	—	—

†All consumers.
*** p<0.001.

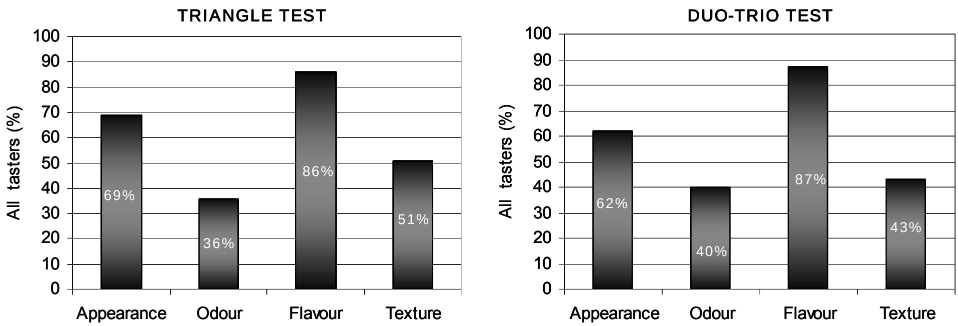

Fig. 1. Attributes indicated by the tasters in the triangle and duo-trio tests for selecting the preferred sample.

Results from the duo-trio test (Table 3) with all consumers pointed out that the consumers ranging 20-60 years significantly differenced the cooked ham manufactured from white or Iberian pork meat. Taking the gender into account, the number of assessors who correctly identified the different sample was lower for male and higher for female. Similarly to triangle test, all tasters chosen the flavour as attribute for differentiating the iberian cooked ham (Fig. 1).

These results may be due to variations in fatty acids content of meat. Different studies have been focused on the role of fatty acids in meat flavour formation In this sense, Mottram (1998) pointed out that the characteristic flavour of cooked meat derives from thermally induced reactions occurring during heating, principally the Maillard reaction and the degradation of lipids. During the oxidation of the fatty acid components of lipids, the reactions occur quickly and provide a profile of volatiles which contribute to desirable flavours. Besides, unsaturated fatty acids undergo autoxidation much more readily than those which are saturated.

Table 3. Results obtained in the duo-trio test carried out with cooked ham

Age	Tasters	No. of correct answers	Significance level
20-30	N†	17/18	***
	Male	9/10	*
	Female	8/8	**
31-40	N	21/23	***
	Male	13/15	**
	Female	8/8	**
41-50	N	25/25	***
	Male	19/19	***
	Female	6/6	*
51-60	N	11/11	***
	Male	10/10	***
	Female	1/1	—
>60	N	5/7	N.s
	Male	5/7	N.s
	Female	—	—

†All consumers.

n.s: $p>0.05$; * $p<0.05$, ** $p<0.01$; *** $p<0.001$.

Finally, the results obtained in the ranking test, showed differences ($p<0.05$) among the three cooked sausages evaluated. The juiciness was the parameter what allowed establish differences. These results revealed that tasters considered Bratwurst sausages as the juiciest and the Iberian sausages as the least juiciness. The fat content of the sausage may explain this result. Matulis *et al.* (1994, 1995) reported that in the manufacture of frankfurters the juiciness differed with the fat content.

IV – Conclusions

In summary, the Iberian cooked meat products evaluated in this study were differentiated by the consumers. These results show that, probably, these types of products can be accepted by the consumer, although studies with a larger number of subjects should be carried out.

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Study of the characteristics of conventional cooked hams and organic cooked hams

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Abstract. Consumers' concern due to the use of additives in the manufacturing of meat product has focused an increasing demand of the organic products. The organic products are primary or manufactured products obtained without the use of chemical products. So, in the organic cooked meat products the use of phosphates is not permitted. However, the use of nitrites is allowed in a lower amount than for manufacturing conventional products due to the role that nitrites exert on the development of colour. The aim of this work was to study the characteristics of conventional and organic commercial cooked hams. Six different commercial brands of cooked hams were analysed: 3 conventional and 3 organics. In each ham the pH, a_w, proximal composition, amount of additives (sodium chloride, nitrites and phosphates) and the colour (L*, a*,b*) were evaluated. The main differences found among the cooked ham analysed were in the protein and fat content. The use of pigs with different genetic or with different feed (conventional or organic) could explain these results. Moreover, differences in the amount of phosphates were detected. The use of different concentration of phosphates in the manufacturing of conventional cooked ham and the use of different raw meats might explain these differences.

Keywords. Additives – Organic products – Cooked ham – Characterization.

Une étude des caractéristiques du jambon cuit conventionnel et du jambon écologique

Résumé. *La préoccupation du consommateur à propos de l'inclusion d'additifs chimiques dans les produits à base de viande a entraîné un développement de la demande de produits écologiques. Les produits écologiques sont des produits agricoles primaires ou élaborés, obtenus sans l'utilisation de produits chimiques. Ainsi, dans les produits à base de viande cuits écologiques, l'ajout de phosphates n'est pas permis. Toutefois, l'inclusion de nitrite est autorisée pour favoriser le développement de la couleur, mais en concentrations plus basses que dans l'élaboration des produits conventionnels. L'objectif de ce travail était d'étudier les caractéristiques du jambon cuit commercial écologique et du jambon cuit commercial conventionnel. Pour cela, 6 marques commerciales de jambon cuit ont été analysées : 3 conventionnelles et 3 écologiques. Pour chaque jambon on a déterminé le pH, l'a_w, la composition proximale, la teneur en additifs (chlorures, nitrites et phosphates) et on a réalisé la mesure instrumentale de la couleur. Les principales différences entre les différents jambons analysés concernaient la teneur en graisse et en protéines. L'utilisation de porcs ayant une génétique différente ou recevant une alimentation différente (conventionnelle ou écologique) pourrait expliquer ces résultats. Des différences ont également été détectées dans la teneur en phosphates. L'emploi de différentes concentrations de phosphates pour l'élaboration de jambons conventionnels et l'utilisation de différentes matières premières pourrait être la cause de ces différences.*

Mots-clés. *Additifs – Produits biologiques – Jambon cuit – Caractérisation.*

I – Introduction

Nowadays, the consumers' demand of additives-free and healthy meat products with quality attributes similar to conventional meat products has focused an increasing demand of the organic products. The organic products are primary or manufactured products obtained without the use of chemical products (DOUE, 2008).

The cooked ham is one of the meat products most demanded in Spain. In the manufacture of this meat product, phosphates (E-450, E-451 and E-452) and nitrites (E-249, E-250) are commonly added. Phosphates improve cohesion of meat pieces and binding of water (Keenan *et al.*, 2010) however, this additive is not permitted for manufacturing organic cooked meat products (DOUE, 2008). Otherwise, the use of nitrites (E-249, E-250) is allowed, due to the role that nitrites exert on the development of colour, but in a lower amount than for manufacturing conventional products. The aim of this work was to study the physico-chemical characteristics of conventional and organic commercial cooked hams.

II – Material and methods

Six different commercial brands of cooked hams were analysed: 3 conventional (C1, C2, C3) and 3 organics (O1, O2, O3). In all cooked hams evaluated (3 for each commercial brand), different analysis were carried out. The pH value was determined with a Crison 2001 pH meter (Crison Instrument S.A, Barcelona, Spain) equipped with a punction electrode. Water activity (a_w) was determined using a hygrometer (Aqua-lab CX2, Decagon, Washington, USA). Moisture, protein, fat and ash content were determined according to ISO standards 1442:1997 (ISO, 1997), 937:1978 (1978), 1443:1973 (ISO, 1973) and 936:1998 (ISO, 1998), respectively. Moreover, NaCl (ISO 1841-1:1996), nitrite (ISO 2918:1975) and phosphate content (ammonium molybdate method, BOE, 1995) were analysed. Surface colour was measured using a reflectance spectrophotometer (CM-2600d/2500d, Konica Minolta, Aquateknia S.A., Spain). Colour coordinates were determined in the CIE-LAB system and the results were expressed as lightness (L*), redness (a*) and yellowness (b*). Data sets were statistically analyzed using one-way variance analysis (ANOVA) in order to determine any significant differences. The means were separated by Tukey-honest significant difference test at 5% level. Principal component analysis (PCA) was performed in order to evaluate the influence of the parameters on total variability. Data analyses were conducted using Statgraphics Plus 4.0 statistical package.

III – Results and discussion

The results obtained for pH, a_w and proximal composition are showed in Table 1. Although one of the organic cooked ham brands presented an unusual pH value according to Aymerich *et al.*, (2003), the pH values for the other brands were similar. For a_w, no differences ($p > 0.05$) were detected among the cooked ham brands evaluated. In general, the values obtained for this parameter were similar than those found by Rubio *et al.*, (2009) in cooked ham.

Table 1. pH, a_w and proximal composition in cooked ham (conventional and organic)

		pH	a_w	Moisture (%)	Protein (%)	Fat (%)	Ash (%)
Conventional	C1	^{b}6.2	0.972	73.3	^{b}18.9	^{ab}2.9	3.3
	C2	^{b}6.5	0.982	73.4	^{b}18.6	^{ab}3.6	3.5
	C3	^{b}6.2	0.972	75.1	^{a}17.1	^{ab}5.0	4.3
Organic	O1	^{b}6.2	0.973	74.4	^{a}17.5	^{b}4.6	3.2
	O2	^{a}5.1	0.973	73.1	^{b}19.0	^{ab}2.1	2.9
	O3	^{ab}5.6	0.979	74.9	^{c}20.5	^{a}1.4	2.8

^{a,b,c} Means within the same column with different letters differ significantly ($p<0.05$).

With regard to proximal composition, differences for the protein and fat content ($p<0.05$) were found among the commercial brands analysed. A higher variability was observed for organic cooked ham in these parameters. The use of raw meat from pigs with different genetic, fed with different diets or slaughtered at different age could affect the composition of raw meat and consequently the composition of meat products (García-Rey *et al.*, 2004). Authors as Cheng, *et al.* (2005) indicated that cooked ham has a moisture, protein and fat content around 65-71%, 21-24% and 1-2%, respectively.

In the Table 2 are showed the results obtained for the content of additives. No differences ($p > 0.05$) were detected for the amount of NaCl. For the nitrite content, it is important to point out that although the maximum amount of nitrites allowed for manufacturing conventional and organic products is different (150 ppm and 80 ppm, respectively), no differences ($p>0.05$) were found among brands since nitrite is a highly reactive compound at the pH of 5.5-6 (Marco*, et al.,* 2006). As it was expected, some differences were found for the phosphate content ($p<0.05$) between conventional and organic brands of cooked ham. The conventional cooked hams showed the highest values for this parameter. The intrinsic value of phosphates that meat has is around 4500 and 5000 ppm (Flores, 2001) and, in Spain, the maximum amount of phosphate permitted is 5000 ppm. Therefore, the use of different concentration of phosphates in the manufacturing of cooked hams might explain the differences found in this study. On the other hand, taking into account that in the manufacture of organic cooked ham the addition of phosphate is not allowed, the differences among the three commercial organic brands may be due to the use of different raw meat.

Table 2. Additives content and colour values obtained in cooked ham (conventional and organic)

		NaCl (%)	Nitrites (ppm)	Phosphates (ppm)	L*	a*	b*
Conventional	C1	1.7	2.1	^{c}7058	65.9	7.6	^{b}9.0
	C2	2.0	3.3	^{a}5018	59.8	8.1	^{a}5.8
	C3	2.0	1.5	^{d}9742	60.4	10.1	^{ab}6.5
Organic	O1	2.1	3.3	^{a}5087	59.8	9.9	^{ab}8.0
	O2	1.4	5.5	^{a}4968	63.4	9.1	^{ab}7.8
	O3	1.7	4.6	^{b}5772	54.6	12.1	^{b}8.6

$^{a,b,c,\,d}$ Means within the same column with different letters differ significantly ($p<0.05$).

Considering colour, only were found differences for b* ($p<0.05$) among the conventional commercial cooked hams. The use of different sorts of ingredients and additives in the manufacture of this product or even the use of different raw material, could explain these differences. The L*, a* and b* values were within the range reported by the others authors with regard to this product (Casiraghi *et al.,* 2007)

Finally, the parameters evaluated were subjected to PCA in order to determine the relationships between the different cooked hams. The plot of the two first principal components (Fig 1) explained 71.78% of the total variance. The first component (PC1) explained 47.92% of the variance. The loading plot for PC1 showed that a_w, a* values, protein and nitrite had a positive loading, whereas the rest of the parameters evaluated had a negative loading. The second PC (PC2) described 23.6% of the variance, where only L* and b* values and phosphate content had a positive loading. The samples distribution on the two first PC plot did not allow clearly separate the two groups. However, differences between them became apparent with respect to PC1. Conventional cooked hams were located on the negative side of the PC1, whereas organic cooked hams were grouped on the positive loading of the PC1. The conventional cooked hams may be distinguished by its phosphate content.

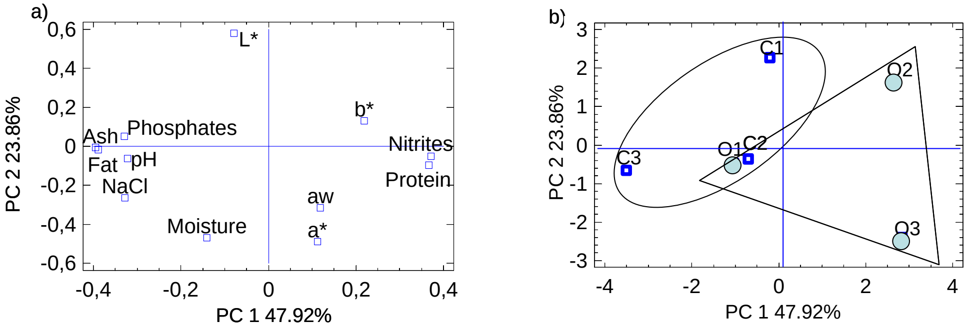

Fig. 1. Loading plot (a) and score plot (b) after principal component analysis of characteristic of cooked ham analysed, conventional (C1,C2,C3) and organic (O1,O2,O3).

IV – Conclusions

On basis of these results, the physico-chemical parameters evaluated did not allow establish clear differences between conventional and organic commercial cooked hams.

Acknowledgments

This material is based upon work (project PEP. 2006.001934) supported by ITACyL and by FEDER.

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Gochu Asturcelta semiextensive production: Carcass and meat quality and fatty acid profile

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Abstract. The breeders association of Gochu Asturcelta (ACGA), has recovered Asturcelta pig breed. The aim of this work is to characterize the carcass and meat produced. Sixteen carcasses were used and carcass weight, dressing percentage and fatness were assessed. Likewise, meat quality parameters as pH, colour (L*, a*, b*), proximal chemical composition, fatty acid profile, water holding capacity, instrumental texture (Warner-Braztler) and sensory analysis were performed. Average carcass weight and dressing percentage were 180 ± 30 kg and 79 ± 3% respectively. Thickness subcutaneous fat was higher than common modern breeds (44 ± 9 mm 6th and 54 ± 6 mm *gluteus medius*). Meat colour measurements revealed lower L* and higher a* values than common commercial breeds. Chemical composition revealed a high intramuscular fat content (9.2 ± 3.5%FM), but fatty acid profile showed a higher content in MUFA than common modern breeds. Water holding capacity was higher than commercial hybrids. Instrumental texture and sensory analysis show that this meat could be defined as tender, juicy and with good palatability.

Keywords. Pork – Carcass – Meat – Fatty acids.

Production semi-extensive du Gochu Asturcelta : Qualité de la carcasse et de la viande et profil en acides gras

Résumé. *L'association des éleveurs Gochu Asturcelta a repris l'élevage de porc Asturcelta. L'objectif de ce travail est de caractériser la carcasse et la viande de ce produit. Seize carcasses ont été utilisées et le poids de la carcasse, le rendement vrai et l'engraissement ont été établis. De même, les paramètres de qualité de la viande comme le pH, la couleur (L*,a*,b*), la composition chimique proximale, le profil en acides gras, la capacité de rétention d'eau, la texture instrumentale (Warner-Bratzler) et l'analyse sensorielle ont été étudiés. Le poids de la carcasse et le rendement vrai étaient de 180 ± 30 kg et de 79 ± 3%. L'épaisseur de la graisse sous-cutanée était plus élevée que chez les races commerciales plus communes (44 ± 9 mm 6th et 54 ± 6 mm sur le* Gluteus medius*). La mesure de la couleur de la viande a révélé un L* plus faible et a* plus élevé que chez les races commerciales. La composition chimique a montré une graisse intramusculaire élevée (9,2 ± 3,5 %) et le profil en acides gras a montré une quantité plus élevée de MUFA que la moyenne. La capacité de rétention d'eau était plus élevée que chez les races commerciales. La texture instrumentale et sensorielle montre que la viande peut être définie comme tendre, juteuse et savoureuse.*

Mots-clés. *Porc – Carcasse – Viande – Profil en acides gras.*

I – Introduction

The changes at world meat markets over the past decade and the improvement in the educational and economical conditions of most consumers have increased the demands in meat they consume. As a consequence, consumers are searching for meat that has characteristics that differ from the most commonly consumed meat. Today, consumers are better informed and more concerned about genetic reserve, production systems and the environment and animal welfare requirements (Fortina *et al.*, 2005; Meinert *et al.*, 2008).

Under this framework, the breeders association of Gochu Asturcelta (ACGA), supported by Principado de Asturias, has recovered Asturcelta pig breed, through a genetic selective program carried out by SERIDA. This Spanish local breed, originated from ancient Celtic breed, characterized by medium-sized and rusticity, is located in the North of Spain. In this area, the natural constraints from the climate and the geography seem to be unfavourable to mass meat production at low cost; consequently, the use of natural sources could be profitable to rear these animals. To produce a high quality meat, however, it is necessary to evaluate variables related to animal production, such as genetic and management properties, as well as variables associated with the processing of meat. In this sense, breed is an important factor that might influence the characteristics of the finished product; therefore, it is presumable that carcass and meat obtained could be different form the most common pork commercialised. Thus, this meat could become an appreciated product that could be sold at high prices in specialty markets and restaurants, and could be considered a profitable alternative for achieving the objective of sustainable meat production, meeting at the same time the European Union requirements about production system, extensification and territory use to contribute to the livelihood of rural population. Te aim of this work was to describe the carcass and meat characteristics of Gochu Asturcelta breed.

II – Materials and methods

Sixteen carcasses of Gochu Asturcelta breed were used. Carcass weight, dressing percentage and backfat thickness (6th rib and M. *gluteus medius* level) were assessed. After the carcasses were cooled for 24 h at 4ºC, the M. *longissimus thoracis* muscle between the 6th and 11th ribs was removed from the left carcass side and divided in portions to analyze meat quality.

In this sense, the following meat quality parameters were analyzed: pH, muscle colour (L*, a*, b*) using the m. *longissimus thoracis* at the 6th rib, proximal chemical composition (moisture, protein and fat content), fatty acid profile, water holding capacity (cooking losses) and instrumental texture (Warner-Braztler). Likewise, in order to perform sensory analysis (descriptive profile), loins were sliced in steaks about 2 cm thick and cooked in double-plate grill (preheated at 220ºC for 10 minutes) to an internal temperature of 70ºC. After cooking, each steak was wrapped in aluminium foil and kept hot until the time of assessment. Using a increasing 5-points scale, a trained eight- member sensory panel was asked for the following sensory characteristics: fresh colour, cooked colour, odour liking, tenderness, juiciness, flavour liking and overall liking.

Results are shown as means and standard deviations for each parameter.

III – Results and discussion

Carcass weight, carcass yield, thickness subcutaneous fat and ultimate pH are reported in Table 1. Average carcass weight and dressing percentage agreed to age at slaughter (14-18 months), and are within values commonly observed when modern hybrids are slaughtered at this age.

Table 1: Carcass characteristics of Gochu Asturcelta breed

Parameter	Average	Standard deviation
Carcass weight (kg)	180.4	30.2
Carcass yield (%)	79.4	3.4
Backfat thickness, 6ª rib level (mm)	44.0	9.0
Backfat thickness, *G. medius* (mm)	54.6	6.0
pH24 M. *longissimus thoracis*	5.78	0.17

Backfat thickness measurements were more closely related with Iberian pig values than with common commercial hybrids (Ramírez and Cava, 2007).

Regarding pH values, although average values were in the normal range, some animals showed pH values closed to the threshold of DFD meat, thus, the values were above those observed in commercial hybrids, but this values are in the line of other studies with local European breeds, who proposed an effect of muscular fibre type and an incomplete post mortem glicolisis, caused by a large of number of genetic factors, as hyposthesis to explain such results (Fortina *et al.*, 2005; Ryu *et al.*, 2008).

Table 2 summarizes nutritional characteristics of meat from Gochu Asturcelta. Intramuscular fat content was higher than average values observed in commercial hybrids. These data, even slightly higher than those obtained in other rustic local breeds (Fortina *et al.*, 2005; Gil *et al.*, 2008; Ryu *et al.*, 2008) confirm that these breeds are prone to adipogenesis.

Table 2: Moisture, fat, protein and fatty acid profile (summary) of Gochu Asturcelta meat

Parameter	Average	Standard deviation
Moisture (%)	69.1	2.9
Fat content (% FM)	9.2	3.5
Protein content (% FM)	21.4	0.9
Saturated fatty acid (%)	36.8	1.1
Monounsaturated fatty acid (%)	55.9	1.5
Polyunsaturated fatty acid (%)	7.3	1.1

Regarding fatty acid composition, several research studies involving different local Italian breeds (Fortina *et al.*, 2005; Pugliese, 2004; Meinert *et al.*, 2008) reported higher saturated, lower monounsaturated and higher polyunsaturated fatty acid percentages than those observed in our trial. These results could be explained by the different feeding system used. In that regard, in mentioned studies a common diet based on concentrates were offered to animals, whilst, in the present study, the feeding strategy included also concentrates but free access to roughage and some fruits such chestnuts. On the other hand, the lowest content in polyunsaturated fatty acid could be caused by the dilution effect of phospholipids rich in these fatty acids due to a high content of intramuscular fat.

The meat quality instrumental parameters and also the scores of sensory parameters given to Astrucelta pig meat are reported in Table 3. Lightness (L*), redness (a*) and yellowness (b*) indicate that Asturcelta pig breed provide darker, redder and yellowier meat than commercial breeds such as Landrace, or Large White. In this sense, Gil *et al.,* (2008) reported values ranged 46.0-48.0 for L*, 2.7-3.0 for a* and 3.6-4.3 for b* for these breeds. Likewise, water holding capacity measured as cooking losses was higher than commercial hybrids. This result could be related with the high content of intramuscular fat, as has been reported in other studies comparing different pig breeds (Pugliese *et al.*, 2004; Gil *et al.*, 2008; Meinert *et al.,* 2008)

Regarding Warner-Braztler shear force, values obtained corresponded with tender meat, although the meat studied was not aged, in consonance with the common practice in the pork industry, because only a slight improvement of tenderness is obtained after ageing in pork meat (Meinert *et al.,* 2008). However, our shear force values are lower than those observed in other studies (Pugliese *et al.*, 2004; Meinert *et al.,* 2008). In this sense, several factors are known to affect tenderness, such as connective tissue, intramuscular fat, which have a major impact (Wood *et al.*, 2004; Meinert *et al.*, 2008; Crawford *et al.*, 2010), thus, the higher content of intramuscular fat observed in our study, could have led to more tender meat.

Sensory evaluation provided medium to high scores for the majority of attributes assessed. The raw and cooked meat colour was scored as darker respect to conventionally commercial pork meat, which is consistent with colorimetric measurements. The scores given to pork meat studied (around 3.5 points in a 5 points scale), indicated that Asturcelta pig breed provides meat with sensorial values slightly above respect to the average.

Table 3. Colorimetric parameters (L*, a*. b*), instrumental texture (Warner-Braztler test) water holding capacity (cooking loses) and sensorial analysis (5 points scale) of Gochu Asturcelta meat

Parameter	Average	Standard deviation
Lightness (L*)	45.9	4.6
Redness index (a*)	6.7	1.7
Yellowness index (b*)	9.0	1.8
Warner-Braztler (kg)	4.3	1.4
Cooking loses (%)	15.2	3.2
Sensorial analysis		
Colour intensity (raw meat)	3.9	0.7
Colour intensity (cooked meat)	3.0	0.7
Odour liking	3.4	0.4
Tenderness	3.2	0.6
Juiciness	3.2	0.5
Flavour liking	3.6	0.3
Overall liking	3.3	0.4

IV – Conclusions

Asturcelta pig breed provide heavier and fatter carcasses than commercial breeds. Meat obtained is characterized by high fat content, with higher percentage of monounsaturated fatty acids than commercial breeds. Instrumental measurements and sensory analysis, show that this meat could be defined as tender, juicy and with good palatability.

Acknowledgments

The Authors want to thank ACGA for giving the data and to SERIDA for the information related to animal production**.** The research was supported by Consejeria de Medio Rural y Pesca (Principado de Asturias) and for the Minsitry of Medio Ambiente y Medio Rural y Marino.

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Carcass performance of fresh meat pieces: sirloin, "presa" and "secreto" in Iberian pig finished at montanera

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Abstract. Cured products (hams, shoulders and loins) from Iberian pig finished at montanera (free-range with diet based on grass and *Quercus* acorns) rise a high value in the market. Nevertheless other pieces are becoming very valuated by consumer of fresh meat. 191 Iberian pigs (Silvela variety) with an average weigh of 163.87± 11.47 kg at slaughter (average carcass weigh = 133,95± 9,83 kg) have been studied to obtain information about three fresh meat pieces: sirloin (*m. Iliopsoas,* and *m. Psoas minor* muscles), "secreto" (*m. Latissimus dorsi* and *m. Trapezius pars cervicalis* muscles) and "presa" (*m. Serratus ventralis thoracis* and *M. Serratus ventralis cervicis* muscles). The weighs and percentage of carcass of these pieces are: sirloins (0.61±0.01 kg and 0.46%± 0.004), "presa" (1.14 ±0.01 kg and 0.85%±0.009) and "secreto" (0.44±0.01 kg and 0.33%±0.003)

Keywords. Iberian pig – Montanera – Carcass – Sirloin – Presa – Secreto.

Rendement de pièces de haute valeur économique chez le porc Ibérique de montanera: le filet, la "presa" et le "secreto"

Résumé. *En plus des pièces qu'on prépare traditionnellement par maturation (le jambon sec, l'épaule sèche et le dos sec), il y a d'autres pièces d'un intérêt spécial chez le porc Ibérique de montanera, dû au prix et au prestige qu'elles atteignent sur le marché. 191 porcs Ibériques de la souche Silvela d'un poids moyen de 163,87± 11,47 kg à l´abattage (poids moyen de la carcasse = 133,95± 9,83 kg) ont été utilisés dans ce travail, qui décrit les performances de trois de ces pièces: le filet (m.* Iliopsoas, *m.* Psoas minor*), le "secreto" (m.* Latissimus dorsi *et m.* Trapezius pars cervicalis*) et la "presa" (m.* Serratus ventralis thoracis *et m.* Serratus ventralis cervicis*). Les résultats nous indiquent que les poids et les performances de la carcasse de porc Ibérique engraissé en montanera sont les suivants: pour les filets (0,61±0,01 kg et 0,46%± 0,004), pour la "presa" (1,14 ±0,01 kg et 0,85%±0,009) et dans le cas du "secreto" (0,44±0,01 kg et 0,33%±0,003).*

Mots-clés. *Porc Ibérique – Montanera – Carcasse – Filet – Presa – Secreto.*

I – Introduction

Iberian pigs fattened in *montanera* (free-range fattening phase with diet based on *Quercus* acorns and grass) produce the most recognized quality products from the *dehesa* (grasslands on cleared Mediterranean forest) and these rise a high value in the market by virtue of it characteristic flavour and their high content in unsaturated fats, as a healthy food.

The most studied and known pieces are those for cured products: hams, shoulders and loins. Nevertheless other pieces are becoming very valuated by consumer of fresh meat (Forero Vizcaíno, 2002; Gómez-Nieves and Robina, 2003). However, traditionally during the slaughtering only sirloins were cut to be sold as a pork piece and others were included in the meat to make traditional Spanish spice-cured sausages, with a lower price in market. Consequently, in order to have a better known and to prevent fraud to consumer, it is important to describe these particular pieces that are being commercialized as fresh meat.

This study is about three of this fresh meat pieces: sirloin (M. *Iliopsoas*, and M. *Psoas minor* muscles), "secreto" (M. *Latissimus dorsi* and M. *Trapezius pars cervicalis* muscles) and "presa" (M. *Serratus ventralis thoracis* and M. *Serratus ventralis cervicis* muscles). These three pieces have been chosen because their increasing interest for consumers and their price in markets (around 15 €/kg). A better known of Iberian pig carcass should contribute to increase its commercial value.

II – Materials and methods

1. Animals

This study was conducted with 120 purebred Iberian pigs (males and females) of the Silvela variety fattened in a *dehesa* of evergreen oaks (*Quercus ilex rotundifolia*) and sacrificed with 163.87± 11.47 kg after ≥ 2 months of *montanera*. All pigs were castrated following the Spanish regulations, to work with the same kind of pigs of the traditional *montanera* system.

After slaughtering the carcasses were stored to chill them at 4ºC during 2 hours to cut these later following the traditional system for Iberian pig.

2. Measures and analysis

All the pieces from the carcasses were weight in a scale with a precision of ±5 g. The results and values in this study are averages and percentages ot the addition of the two pieces of each animal.

SPPS 11.5© was used for statistical analysis (mean ± standard error) and Pearson correlations.

III – Results and discussion

Table 1 shows the carcasses performance with an average weight of 133.95± 9.83 kg. The values of weigh and percentages of sirloin, *"presa"* and *"secreto"* were respectively: 0.61±0.01 kg and 0.46%± 0.004, 1.14 ±0.01 kg and 0.85%±0.009, and 0.44±0.01 kg and 0.33%±0.003. The three pieces together represent 1.64±0.012 % of the carcass weight.

Table 1. Weigh and percentages of sirloins, *presas* and *secretos* (two from every carcass)

		Carcass		Sirloins		*"Presas"*		*"Secretos"*		Sirloins, *"presas"* and *"secretos"*
		(kg)	(%)	(kg)	(%)	(kg)	(%)	(kg)	(%)	(%)
Mean		133.95	81.53	0.61	0.46	1.14	0.85	0.44	0.33	1.64
Standard error		0.71	0.16	0.01	0.004	0.01	0.009	0.01	0.003	0.012
Minimum value		111.80	76.99	0.40	0.28	0.87	0.66	0.23	0.19	1.24
Maximum value		197.00	89.62	0.89	0.65	2.13	1.67	0.70	0.50	2.45
Percentiles	25	127.30	80.4238	0.56	0.42	1.05	0.79	0.40	0.30	1.55
	50	133.70	81.2607	0.60	0.46	1.13	0.84	0.45	0.33	1.63
	75	139.30	82.4608	0.65	0.49	1.20	0.90	0.50	0.36	1.73

Table 2 shows the Pearson correlations. The weighs of the three pieces are inversely correlated (P< 0.01). However, although these three pieces weighs are positively correlated with carcass weight, the correlation between the carcass percentage of these pieces and the carcass weight is negative.

Table 2. Pearson correlation for carcass weight and performance and different pieces

	(1)	(2)	(3)	(4)	(5)	(6)	(7)	(8)
(1) Carcass percentage	0.06							
(2) Sirloins (kg)	0.32 **	0.16						
(3) *"Presas"* (kg)	0.21 **	0.13	0.24 **					
(4) *"Secretos"* (kg)	0.41 **	-0.21 *	0.27 **	0.12				
(5) Sirloins (%)	-0.19 **	0.15	0.86 **	0.13	0.06			
(6) *"Presas"* (%)	-0.28 **	0.10	0.06	0.88 **	-0.09	0.20 **		
(7) *"Secretos"* (%)	-0.05	-0.25 **	0.13	0.02	0.89 **	0.15 *	0.04	
(8) Sirloins. *"presas"* and *"secretos"* (%)	-0.30 **	0.05	0.40 **	0.74 **	0.22 **	0.57 **	0.87 **	0.39 **

(**) P< 0.01; (*) p< 0.05.

IV – Conclusions

Sirloins. *presas* and *secretos* from Iberian pigs represent a low percentage of carcass. Hence. having in mind that these are becoming more popular and reach a higher price than other pork products, these should be well described to avoid fraud to consumers.

Acknowledgements

The authors wish to acknowledge to Turcañada S. L. and Camilo Rios S.L their collaboration.

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Leaf lard and backfat thickness relation at slaughter in pure breed Iberian pigs finished at montanera

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Abstract. Leaf lards (fat deposit surrounding the kidneys) is the most valuated fat from Iberian pig. 120 pure breed Iberian pigs, Silvela variety, finished at *montanera* (free-range with diet based on grass and *Quercus* acorns) with mean slaughter weigh of 162.53 ± 1.71 kg have been studied to know if it possible to relate leaf lards weigh (llw = 5.15± 0.05 kg) with backfat thickness (bf), measured at two different levels: last thoratic vertebra level (bflt = 6.67± 0.06 cm) and fourth lumbar vertebra level (bf4l = 9.45±0.08). Linear regression models for each level (llw = 2.36+0.46*bflt) and (llw = 6.24+0.117*bf4l) have a low R^2 value, hence bf thickness is not related with llw, and consequently these fattening measures are not reliable to estimate leaf lards weigh.

Keywords. Iberian pig – Montanera – Back fat – Leaf lard.

Rapport entre le poids des crépines (tissu adipeux périrénal) et l'épaisseur de graisse dorsale à l'abattage chez le porc Ibérique à finition montanera

Résumé. *La graisse extraite des crépines (tissu adipeux périrénal) est la graisse la plus estimée de celles qu'on obtient du porc Ibérique. À partir de 120 porcs Ibériques de la souche Silvela à finition montanera, avec un poids moyen à l´abattage de 162,53 ± 1,71 kg, on étudie le rapport possible entre le poids des crépines (llw = 5,15± 0,05 kg) et l'épaisseur de graisse dorsale (bf) au niveau de la dernière vertèbre thoracique (bflt = 6,67± 0,06 cm) et de la quatrième vertèbre lombaire (bf4l = 9,45±0,08). Les modèles de régression obtenus pour chacun des niveaux: vertèbre thoracique (llw = 2,36+0,46*bflt) et quatrième vertèbre lombaire (llw = 6,24+0,117*Bf4l) ne sont pas utiles pour estimer d'une façon fiable llw à partir du bf, parce qu'elles présentent toutes les deux une valeur de* R^2 *très faible. Par conséquent, l'épaisseur de graisse dorsale comme mesure d'engraissement n'a pas de rapport avec le poids des crépines.*

Mots-clés. *Porc Ibérique – Montanera – Crépines – Graisse dorsale.*

I – Introduction

Iberian pigs fattened in *montanera* (free-range fattening phase with diet based on *Quercus* acorns and grass) produce the most recognized quality products from the *dehesa* (grasslands on cleared Mediterranean forest). Cured hams and shoulders obtained from these free-range pigs have gained widespread consumer acceptance and a high commercial value by virtue of it characteristic flavour; also, the high content in unsaturated fats of the ham has increased its appreciation as a healthy food. This rearing regime determines the fatty acid composition of pig fat in terms of four main fatty acids: oleic, linoleic, palmitic and stearic (Alonso *et al.*, 2008).

This production is geographically reduced to the South West of the Iberian Peninsula, and mast of acorns is limited to fall and winter seasons. The leaf lard from these pigs is the most estimated lard in the market (Forero Vizcaíno, 2002).

In this study it has been analyzed the relation between the back fact thickness and the weight of the leaf lards in Iberian pigs in order to know if the back fact thickness could predict the weight of the leaf lards, as method of measuring the greasing degree of the carcasses (Edwards *et al*., 1992; Medel and Fuentetaja, 2000).

II – Materials and methods

This study was conducted at a *dehesa* of evergreen oaks (*Quercus ilex rotundifolia*) with 120 purebred Iberian fattening pigs (male and female) of the Silvela variety. Pigs were on average 111.8±0.7 kg of LW at the start of the study and 162.53 ± 1.71 kg at the end, after ≥ 2 months (69.90 ± 0.45 days). All pigs were castrated following the Spanish regulations, to work with the same kind of pigs of the traditional *montanera* system. The stocking rate (0.76 pigs/ha) was established with margins that guaranteed that the acorns would not run out before the fattening was completed (Rodríguez-Estévez *et al*., 2007, 2008). The carcasses were cut according Iberian pig traditional pork industry.

The leaf lards were weighed individually after slaughtering, and the backfat thickness was measured at two different levels: last thoratic and fourth lumbar vertebra level.

SPPS 11.5© was used for statistical analysis (mean ± standard error) and linear regression models.

III – Results and discussion

The leaf lards weighed 5.15± 0.05 kg and the back fat thicknesses were: 6.67± 0.06 cm at last thoracic vertebra level and 9.45±0.08 cm at fourth lumbar vertebra level.

Table 1 shows the linear regression models for each level; according to the low R^2 values these are not suitable for predictions. Figure 1 represents the weigh of leaf lards and the back fat thicknesses and it is possible to observe the data dispersion.

Table 1. Linear regression models for leaf lard weighs

Independent variable	Linear regression	R^2	Standar Error	Sig.
Back fat thickness at last thoracic vertebra level (bflt)	= 2.36-0.46*bflt	0.342	0.709	0.000
Back fat thickness at fourth lumbar vertebra level (bf4l)	= 6.23-0.117*bf4l	0.034	0.69878	0.013

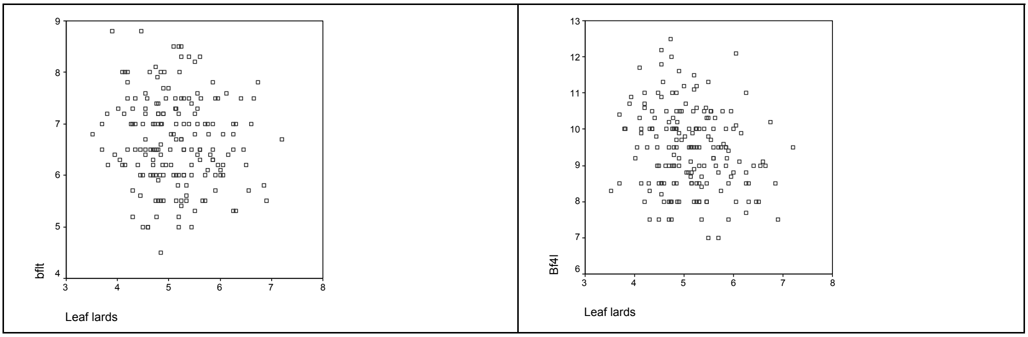

Fig. 1. Regression graphics of leaf lards weighs (kg) and back thickness (cm) at last thoracic vertebra level (bflt) and fourth lumbar vertebra level (bf4l).

IV – Conclusions

The backfact thickness measures at different levels as a fattening measure are not reliable to estimate leaf lard weighs in Iberian pig.

Acknowledgements

The authors wish to acknowledge to Turcañada S. L. and Camilo Rios S.L. their collaboration.

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Lipids performance of biopsies from the subcutaneous back tissue of Iberian pig along montanera fattening period

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Abstract. Iberian pigs fattened at *montanera* (free-range fattening phase, with diet based on grass and *Quercus* acorns) products are the most recognized from *dehesa*, because their high contents in unsaturated fatty acids. Spanish regulation establishes an official method to separate commercial categories depending of fattening period feed. Fatty acid profile evolution during fattening period helps stockmen to organize grazing and to decide slaughter date. In this study 120 pure breed Iberian pigs have been studied using the "Spring loaded biopsy" method for *in vivo* biopsy; obtaining 1.45 ± 0.28 g of tissue at the beginning of *montanera* (1st day) and 1.26 ± 0.68 g one month later. Fat was extracted by microwave to obtain 0.83 ± 0.17 g and 0.73 ± 0.23 g from the first and second biopsy respectively. A regression model has been obtained to predict the amount of extracted fat knowing biopsies weigh (EF=-0.218+0.767*BW first biopsy and EF=0.046+533*BW in the second). The amounts of fat obtained are enough for individual fatty profile analysis following the official methods of analysis.

Keywords. Iberian pig – Montanera – Back fat.

Rendement en gras des biopsies sous-cutanées du porc Ibérique en début et après un mois de montanera

Résumé. *Les produits des porcs Ibériques engraissés en montanera sont ceux qui atteignent une valeur commerciale plus élevée, ceci est dû, principalement, à leur teneur en acides gras insaturés. Actuellement, pour déterminer les catégories commerciales, la législation espagnole préconise la méthode de l'analyse du profil en acides gras de la carcasse. La connaissance de l'évolution du profil en acides gras pendant l'engraissement permet d'organiser le pâturage et de programmer les abattages. Dans ce travail, à partir de 120 porcs Ibériques purs, on étudie la quantité de tissu sous-cutané obtenu grâce à la méthode de biopsie* in vivo *"Spring loaded biopsy" (1,45 ± 0,28 g à l'arrivée en montanera et 1,26 ± 0,68 g après un mois), et la quantité de graisse extraite par micro-ondes (0,83 ± 0,17 g dans la première biopsie, et 0,73 ± 0,23 g dans la deuxième). On propose des modèles de régression qui nous permettent de connaître la quantité de graisse qu'on obtiendrait à partir du poids de la biopsie (EF=-0,218+0,767*BW dans la première biopsie et EF=0,046+0,533*BW dans la deuxième). Les résultats indiquent que ces quantités de graisse sont suffisantes pour l'analyse individuelle du profil en acides gras.*

Mots-clés. *Porc Ibérique – Montanera – Graisse dorsale.*

I – Introduction

The Iberian pig constitutes a breed of great economic importance in Spain and Portugal. In the traditional rearing system, pigs are free-reared in an expanse of land of variable area, using natural resources [mainly grass (*Quercus ilex*) and acorns (*Quercus suber*)] only. Cured ham obtained from free-range pigs has gained widespread consumer acceptance and a high commercial value by virtue of it characteristic flavour; also, the high content in unsaturated fats of the ham has increased its appreciation as a healthy food. The free-range rearing system departs considerably from the intensive farming regime, where pigs are confined and fattened with commercial feed. The Spanish official method for discriminating between pig feeding and rearing regimes determines the fatty acid composition of pig fat in terms of four fatty acids (oleic, linoleic, palmitic and stearic).

The traditional husbandry system of the Iberian pig in the *dehesa* (cleared Mediterranean forest like savannah) is linked to the sustained use of the pasturelands, finishing pigs during the acorn mast-feeding (called *montanera*). Rodríguez-Estévez *et al*. (2009) suggest a daily DM intake of 3.1-3.6 kg acorn kernel and 0.38-0.49 kg grass, which is achieved thanks to the functional characteristics of this breed. Free range Iberian pigs fattened with acorns give the most recognized products from the *dehesa*; however this production is geographically reduced to the South West of the Iberian Peninsula Spain, and acorn production is limited to fall and winter seasons.

Montanera products attain the highest prices in the market because of preference by consumers. The main reason for this preference is the high oleic acid content in the fat of these pigs free-range fattened (Daza *et al*., 2007), mostly due to the high oleic acid content of acorns. This high proportion of oleic acid in the carcass strongly influences the properties of fat, leading a soft and oily lard, which is highly appreciated by consumer, who select these products for special occasions and pays a high price for them (López-Bote, 1998). Moreover, the higher oxidative stability of this pork, because of high α-tocopherol content of grass and γ-tocopherol of acorns improves technological and sensorial quality of Iberian pigs meat and products (Daza *et al*., 2007).

To prevent fraud the Iberian pig Spanish regulations establish an official method to evaluate the finishing diet regime (MAPA, 2007), based on the analysis of the subcutaneous backfat tissue fatty acid profile.

The aim of this study is to know the lipid performance of biopsies from subcutaneous back tissue as source of fat to analyze the fatty acid profile evolution during the *montanera* fattening.

II – Materials and methods

The experimental procedures and animal care conditions were approved by the Animal Experimentation Ethical Committee of the University of Córdoba, Spain.

1. Animals and handling

The study was conducted at a *dehesa* of evergreen oaks (*Quercus ilex rotundifolia*) with 120 purebred Iberian fattening pigs (male and female) of the Silvela variety. Pigs were on average 111.8±0.7 kg of LW at the start of the study and 157.9±1.7 kg at the end, after ≥ 2 months. All pigs were castrated following the Spanish regulations, to work with the same kind of pigs of the traditional montanera system. The stocking rate (0.76 pigs/ha) was established with margins that guaranteed that the acorns would not run out before the fattening was completed (Rodríguez-Estévez, 2007; Rodríguez-Estévez *et al*., 2008)

2. Weighing of the pigs

To calculate body weigh (BW), all the pigs of the herd were weighed individually at the beginning of *montanera*, after a month and the day previous slaughtering. An electronic scale (precision of 100 g) was used for weighing.

3. Biopsy collection

The biopsies were taken the first day of montanera and one month later. The samples were collected *in vivo* with a *Spring loaded biopsy* instrument, following the method described by (Bosch Puig *et al*., 2008). Once the sample was extracted, these samples were collected from the cannula and introduced in individual plastic tubes with the animal reference number, cleaning properly the cannula before taking the next sample.

All biopsies were transported at ≈ 10ºC to the laboratory, where these were stored at –25ºC, until their processing.

4. Lipid extraction

After completely defrosting each sample was weighed before using the technique of microwave oven described by De Pedro *et al*., 1997 to extract the fat from these tissue biopsies. Later the fat obtained from each biopsy was weighed and frozen to store it for future analysis of fatty acid composition.

III – Results and discussion

Descriptive statistics of weights of first and second biopsies (first day and one month later respectively) are showed in Fig. 1 and Table 1.

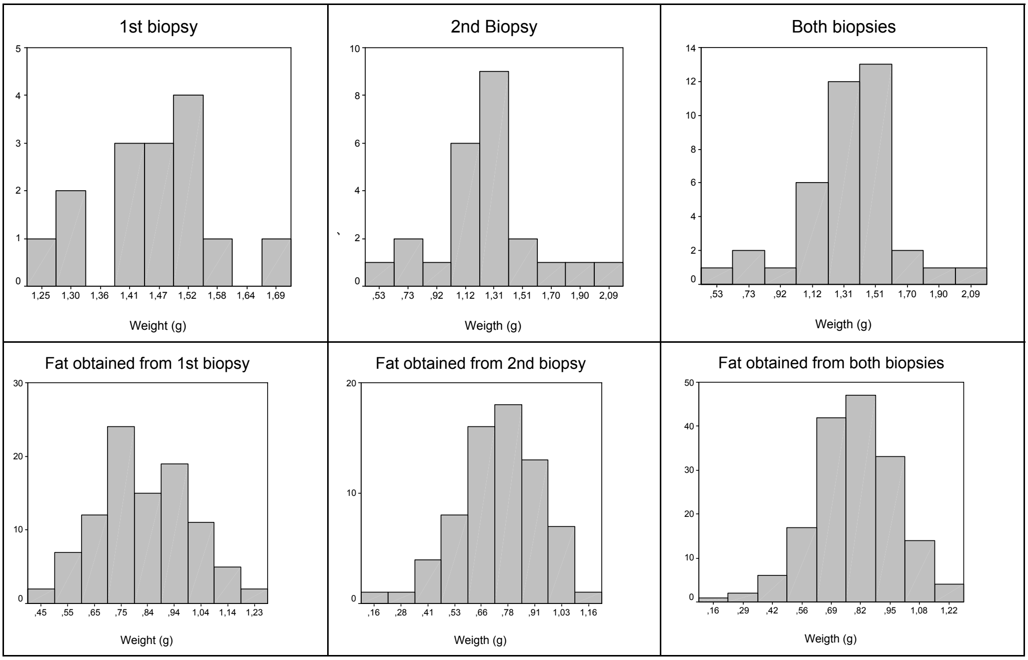

Fig. 1. Weigh of first, second and both biopsies and of fat obtained by microwave extraction.

Table 1. Biopsy weight and fat obtained in the 1st and 2nd biopsies

	1st Biopsy Weight	Fat obtained	2nd Biopsy Weight	Fat obtained	Both Biopsies Weight	Fat obtained
Mean±SE	1.45±0.28	0.83±0.17	1.26±0.68	0.73 ± 0. 23	1.33 ± 0.45	0.79 ± 0. 14
Minimum value	1.27	0.42	0.54	0.15	0.54	0.15
Maximum value	1.67	1.24	2.10	1.17	2.10	1.24
Percentile 25	1.42	0.74	1.08	1.08	1.21	0.68
Percentile 50	1.45	0.83	1.24	1.24	1.34	0.79
Percentile 75	1.52	0.93	1.38	1.38	1.51	0.91

Table 2. Linear regression models for the weight of fat obtained from biopsies (BW = body weight)

Effect variable	Linear regression	R^2	Standard Error	Sig.
Fat obtained (first biopsy)	=-0,218+0,767*BW	0,419	0,102	0,009
Fat obtained (second biopsy)	=0,046+533*BW	0,665	0,128	0,009

As Table 2 shows, it is possible to determine the final fat amount through linear regressions either in the first biopsy (fat obtained = -0,218+0,767*BW) or the second one (fat obtained = 0.046+533*BW).

Results show that it is possible to use this method of fat extraction, because it allows to obtain the necessary amount of fat to determine the fatty acid profile trough different methods of analysis, as it is shown in Table 3.

Table 3. Comparison of different analytical methods to determine the fatty acid profile

Reference	Method of extraction of fat	Method of analysis	Needed amount of fat extracted
(Carrapiso *et al*., 2001)	Dissection	Electronic nose	5 g (adipose tissue)
ISO 15304			250 mg
(Presidencia, 2004) (Oficial method)	Dissection and solvent or microwave extraction.	Gas Chromatography	0.2 g
(Alonso *et al*., 2008)	Not specified	Ion Mobility Spectrometry	1 g
(López-Vidal *et al*., 2008)	Microwave	Gas Chromatography - Mass Spectrometry and Chemometrics	0.2 g
(Arce *et al*., 2009)	Microwave	Infrared spectroscopy	A few microlitres
(Pascual *et al*., 2006)	Folch, Lee and Stanley (1957)	Gas Chromatography	0.5 g
(Regueiro *et al*., 2006)	ISO-1443 (Soxhlet) Folch, Lee and Stanley (1957) Bligh and Dyer (1959) Accelerated extraction methods Hexano-2, propanol 2:1	Gas Chromatography (FID) -Mass Spectrometry and Chemometrics	

IV –Conclusions

The combination of the *Spring loaded biopsy* and microwave fat extraction could be used to predict and to categorize animals before finishing and during the *montanera* in order to determine their handling and their slaughter date. It is possible to obtain *in vivo* biopsies and the amounts of fat obtained are enough for individual fatty acid profile analysis following the official methods of analysis.

Acknowledgements

The authors wish to acknowledge to Turcañada S. L. and Camilo Rios S.L their collaboration.

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Sex influence in carcass (hams and shoulders) performance of Iberian pigs fattened at montanera

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Abstract. Sex factor (castrated males *vs* castrated females) has been studied in relation with montanera fattening (free-range with diet based on grass and *Quercus* acorns) and carcass performance (pieces which will be transformed by dry-curing process: hams, shoulders and loin) of 81 pure Iberian pigs (Silvela variety) with an average slaughter weigh of 167.18± 1.08 kg. The results show that males are more efficient. Results show that fresh hams and shoulders from males before and after cutting are heavier than from females (hams: before 13.80 *vs* 13.29 kg and after cutting 10.78 *vs* 10.43 kg; and shoulders: before 9.40 vs 8.88 kg and after cutting 7.19 *vs* 6.79 kg), and that, after a year of curing process, cured shoulders and hams from males are significantly heavier (cured shoulders: 5.31 *vs* 4.98 kg; cured hams 8.42 *vs* 8.19 kg). By other side dry-curing process produces a significant bigger weight loss in female shoulders (26.73 *vs* 26.14 %) and in male hams (2.41 *vs* 2.30 kg). In conclusion it is of higher interest to reserve montanera pastures to finish preferment males.

Keywords. Iberian pig – Montanera – Ham – Shoulder – Sex.

Différences de rendement selon le sexe pour la carcasse du porc Ibérique de montanera : le jambon et l'épaule

Résumé. *On étudie l'influence du facteur sexe (mâles castrés vs femelles castrées) sur le rendement de carcasses de pièces de haute gamme (jambons secs, épaules sèches et dos secs) sur 81 porcs Ibériques purs engraissés en montanera, de la souche Silvela, dont le poids moyen à l'abattage était de 167,18 ± 1,08 kg. Les résultats indiquent que les jambons et épaules frais provenant des mâles avant et après le découpage pèsent nettement plus que ceux provenant des femelles (jambons: avant le découpage 13,80 vs 13,29 kg, et après 10,78 vs 10,43 kg ; épaules : avant le découpage 9,40 vs 8,88 kg, et après 7,19 vs 6,79 kg). Nous notons la même différence significative selon le sexe à la fin d'une année de vieillissement (jambons : 8,42 vs 8,19 kg ; épaules : 5,31 vs 4,98 kg). Le pourcentage de perte de poids dans le procédé de vieillissement est supérieur chez les femelles pour les épaules (26,73 vs 26,14 %) et chez les mâles pour les jambons (2,41 vs 2,30 kg). Nous en concluons donc qu'il est plus intéressant de réserver la montanera pour engraisser les mâles.*

Mots-clés. *Porc Ibérique – Montanera – Jambon – Épaule – Sexe.*

I – Introduction

Free-range finishing of Iberian pigs based on *Quercus* acorns and grass (called *montanera*) is a strategic production of the *dehesa* (grasslands on cleared Mediterranean forest) in the southwest of the Iberian Peninsula and it is very important for the economy of this agro ecosystem. The study of diferent factors in relation with efficiency is necessary to maximize profits, preserving traditional breeding and quality products (López-Bote, 1998).

Commercial value of Iberian pigs products in markets comes from his traditional breeding system: pure Iberian breed and free-range finishing, which give to their products (hams and shoulders) a high price because their organoleptic properties which make different from others (León Crespo, 1992).

There are different factors with influence on carcass performance: breed and variety, age, feeding regime, husbandry and sex. Traditionally Iberian pigs of both sexes are castrated while these are piglets to maintain pork quality and to avoid handling problems, as attracting wild boars.

This research has been conducted to study the sex factor (castrated males vs castrated females) in relation with carcass performance (hams and shoulders) of pure Iberian pigs (Silvela variety). To avoid other factors with influence, as variety (Clemente *et al*., 2008) or pasture differences (Rodríguez-Estévez *et al*., 2009), all the animals for this experience were of the same variety and were fattened in the same *dehesa* during the same *montanera*.

II – Materials and methods

1. Animals

81 Iberian pigs (pure breed) of the Silvela variety were used (40 castrated males and 41 castrated females) Animals start montanera period with an average of 343 ± 8 days of life and a weigh of 113.5 ± 3.5 kg. All these pigs were bred under the same conditions and were finished during ≥ 60 days of *montanera*, until these gained the minimum traditional slaughter weigh of 161 kg (14 @), with an average slaugther weigh of 167.18± 1.08 kg (Table 1). At the start of *montanera* there were no statistical differences for age and weight between males and females.

During the finishing the animals grazed in a *dehesa* of Fuente Obejuna (Córdoba-Spain) and their diet was based only on *Quercus* acorns grass.

2. Measures and analysis

Both hams and shoulders from every carcass were weighed in a scale with a precision of ±5 g. Weights correspond to: (i) 24 h after slaughter before cutting; (ii) after cutting to prepare pieces for curing process; (iii) after 1 year of curing process. The results presented here correspond to mean, standard error, minimum and maximum of absolute values and percentages of the two pieces of each animal. SPPS® has been used to statistical analysis. Comparisons between males and females have been made by ANOVA.

III – Results and discussion

Males and females showed different weight gain; hence these had different slaughter weight (Table 1) ($P<0.01$). The results show that fresh hams and shoulders from males before and after cutting were heavier than from females (hams: before 13.80 vs 13.29 kg and after cutting 10.78 vs 10.43 kg, $P<0.001$, Table 2; and shoulders: before 9.40 vs 8.88 kg and after cutting 7.19 vs 6.79 kg, $P<0,001$, Table 3). All the same, after a year of curing process, cured hams and shoulders from males were significantly heavier (cured hams 8.42 vs 8.19 kg, $P<0.001$, Table 2; and cured shoulders: 5.31 vs 4.98 kg, $P<0.001$, Table 3). By other side, the dry-curing process gave different weight losses for females and males; so weight losses of hams were higher for males (2.41 vs 2.30 kg, $P<0.05$) and weight losses of shoulders were higher for females (26.73 vs 26.14 %, $P<0.05$).

Table 1. Body weight differences between males and females at slaughter ($P < 0.01$)

Animals	Animals	Mean	Standard error	Minimum	Maximum
Males	40	167.18	1.08	146.10	190.60
Females	41	159.34	1.24	146.40	197.50
Total	81	163.21	0.88	146.10	197.50

Table 2. Weights of hams according to sex

		n	Average	Standard error	Minimum	Maximum
Hams before cutting***	Males	80	13.80	0.11	11.62	16.75
	Females	82	13.29	0.12	11.25	16.85
	Total	162	13.54	0.08	11.25	16.85
% of hams about carcass weight	Males	80	20.44	0.10	18.02	22.11
	Females	82	20.53	0.09	18.63	22.42
	Total	162	20.48	0.07	18.02	22.42
Hams after cutting***	Males	80	10.78	0.09	9.00	13.10
	Females	82	10.43	0.09	8.80	13.25
	Total	162	10.60	0.07	8.80	13.25
% of cut hams about carcass weight	Males	80	15.97	0.10	13.81	17.90
	Females	82	16.12	0.10	14.20	18.16
	Total	162	16.05	0.07	13.81	18.16
Weight after 1 year of curing***	Males	80	8.42	0.08	7.05	10.35
	Females	82	8.19	0.07	6.70	10.55
	Total	162	8.30	0.05	6.70	10.55
Weight loss after 1 year of curing (kg)	Males	80	2.41	0.02	2.00	2.95
	Females	82	2.30	0.03	1.51	3.05
	Total	162	2.35	0.02	1.51	3.05
% weight loss after 1 year of curing*	Males	80	22.41	0.17	17.39	25.98
	Females	82	22.01	0.23	14.60	26.87
	Total	162	22.21	0.14	14.60	26.87

Statistical differences: * for P < 0,05; ** for P < 0,01; *** for P < 0,001.

Table 3. Weights of shoulders according to sex.

		n	Average	Standard error	Minimum	Maximum
Shoulder before cutting***	Males	80	9.40	0.09	7.43	11.42
	Females	82	8.88	0.07	7.85	11.00
	Total	162	9.13	0.06	7.43	11.42
% of shoulders about carcass weight	Males	80	13.91	0.09	12.29	15.93
	Females	82	13.73	0.08	12.38	15.72
	Total	162	13.82	0.06	12.29	15.93
Shoulder after cutting***	Males	80	7.19	0.07	5.95	9.35
	Females	82	6.79	0.07	5.75	8.75
	Total	162	6.99	0.05	5.75	9.35
% of cut shoulders about carcass weight	Males	80	10.64	0.09	9.11	12.99
	Females	82	10.50	0.08	8.96	12.69
	Total	162	10.57	0.06	8.96	12.99
Weight after 1 year of curing***	Males	80	5.31	0.06	4.35	6.90
	Females	82	4.98	0.05	4.20	6.65
	Total	162	5.14	0.04	4.20	6.90
Weight loss after 1 year of curing (kg)	Males	80	1.79	0.09	-5.05	2.45
	Females	82	1.73	0.08	-4.80	2.15
	Total	162	1.76	0.06	-5.05	2.45
% weight loss after 1 year of curing*	Males	80	26.14	0.20	21.92	29.69
	Females	82	26.73	0.18	21.71	30.27
	Total	162	26.44	0.14	21.71	30.27

Statistical differences: * for P < 0,05; ** for P < 0,01; *** for P < 0,001.

IV – Conclusions

In conclusion it is of higher interest to reserve the *montanera* (mast of acorns) preferentially to finish castrated males, because these gain more weight and have a highest carcass performance for the most valuated pieces from Iberian pig: hams and shoulders.

Acknowledgements

The authors wish to acknowledge to Turcañada S. L. and Camilo Rios S.L. their collaboration.

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Slaughter weight, carcass performance and backfat thickness relations with losses during the cutting process of hams and shoulders from Iberian pigs fattened at *montanera*

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Abstract. Cutting process of hams and shoulders from Iberian pigs is based in removing some muscle mass, fat and skin to obtain the traditional and standard commercial shape, which makes these different from other commercial cured pieces. There is a significant correlation ($p<0.01$) between backfat thickness (measured at two different levels: last thoracic vertebra 6.67 ± 0.87 cm and 4th lumbar vertebra 9.45 ± 1.12 cm) and weigh loss in hams and shoulders by cutting process (5.95 ± 1.09 kg and 21.40% ± 3.32 in hams vs 4.30 ± 0.61 kg 23.09 ± 2.77% in shoulders). Besides there is significant correlation ($p<0.05$) between ham weigh losses by cutting process and carcass performance, and between shoulder weigh losses by cutting process and slaughter and carcass weighs ($p<0.01$).

Keywords. Iberian pig – Montanera – Back fat – Slaugther weigh – Carcass performance – Hams – Shoulder – Cutting process.

Corrélations entre le poids à l'abattage, le rendement de la carcasse, l'épaisseur de graisse dorsale et les pertes au découpage pour les jambons et les épaules du porc Ibérique pur de "bellota"

Résumé. *Le processus de découpage consiste à éliminer une part de la musculature, de la graisse et de la peau, pour obtenir une pièce d'une forme et d'une taille précises, selon les standards commerciaux du jambon et de l'épaule Ibériques, ce qui fait qu'il est différent d'autres types de jambon. Il y a une corrélation significative ($p<0,01$) entre le poids perdu dans les jambons et les épaules par découpage (5,95 ± 1,09 kg et 21,40% ± 3,32 dans les jambons vs 4,30 ± 0,61 kg et 23,09 ± 2,77% dans les épaules) et l'épaisseur de la graisse dorsale au niveau de la dernière vertèbre thoracique (6,67 ± 0,87 cm. d'épaisseur) et de la quatrième vertèbre lombaire (9,45 ± 1,12 cm d'épaisseur). D'autre part, dans les jambons on constate une corrélation ($p<0,05$) entre le rendement de la carcasse et les pertes par découpage. Par ailleurs, il existe une corrélation positive ($p<0,01$) entre les pertes par découpage dans les épaules et le poids vif et le rendement et le poids de la carcasse.*

Mots-clés. *Porc Ibérique – Montanera – Graisse dorsale – Poids à l'abattage – Rendement de la carcasse – Jambons – Épaules – Découpage.*

I – Introduction

The Iberian pig constitutes a breed of great economic importance in Spain and Portugal. Cured hams obtained from free-range pigs finished using natural resources (basically *Quercus* acorns and grass) has gained widespread consumer acceptance and a high commercial value by virtue of it characteristic flavour and their high content in unsaturated fats. This production is geographically reduced to the South West of the Iberian Peninsula during the fall and winter seasons.

After slaughtering the carcass, hams and shoulders are cut previously to the dry-curing process during a process called cutting (Forero Vizcaíno, 2002). This is based in removing some muscle, fat and skin in hams and shoulders to obtain the traditional and standard commercial shape, which makes these different from other hams; it mainly is based in a section in V form in muscular and exterior face from the top of the ham until the hoke level (Gómez-Nieves And Robina, 2003)

By other side, the measuring of back fat thickness is the usual method to evaluate the fattening or greasing degree of the carcasses (Edwards *et al*., 1992; Fortin, 1986; Mayoral *et al*., 1999; Medel and Fuentetaja, 2000) and usually it is a complementary measurement with the carcass performance (carcass weigh in relation to animal weigh), in order to have more information about the final performance and rentability of different husbandry systems, varieties, strains and genetical lines of Iberian pigs (Ellis *et al*., 1996; Latorre *et al*., 2003).

This research has been conducted to know if there is any relation between the carcass performance and the backfat thickness with the weigh of losses during the cutting process of shoulders and hams from the Iberian pig carcass.

II – Materials and methods

191 Iberian pigs (males and females) of the Silvela variety fattened in *montanera* were used. The animals were slaughtered after 69.90 ± 0.45 days eating only acorns and grass, with a mean weigh of 162.53 ± 1.71 kg according to Spanish regulations and standard commercial procedures. All pigs were castrated following the Spanish regulations, to work with the same kind of pigs of the traditional montanera system. The stocking rate (0.76 pigs/ha) was established with margins that guaranteed that the acorns would not run out before the fattening was completed (Rodríguez-Estévez *et al*., 2008).

The backfat thickness was measured at two levels: last thoracic vertebra and fourth lumbar vertebra level. Hams and shoulders were individually weighed before and after the cutting process; All the pieces from the carcasses were weight in a scale with a precision of ±5 g. The results and values in this study are averages and percentages for the addition of the two pieces of each animal.

SPPS 11.5© was used for statistical analysis (mean ± standard error) and to model a linear regression.

III –Results and discussion

The backfat thicknesses measured at the two levels were: 6.67±0.87 cm at the last thoracic vertebra and 9.45±1.12 cm at the fourth lumbar vertebra level. Table 1 shows hams and shoulders weighs and percentages before and after cutting process.

Table 1. Hams and shoulders weighs and percentages before and after cutting process

	Minimum value	Maximum value	Mean	Standard error
Fresh shoulders before cutting (kg)	14.90	24.00	18.81	1.46
Fresh hams before cutting (kg)	13.00	34.65	29.35	3.79
Fresh shoulders after cutting (kg)	11.10	18.40	14.33	1.25
Fresh hams after cutting (kg)	17.70	26.15	21.75	1.68
Weigh losses in hams (kg)	0.15	9.55	5.95	1.09
Weigh losses in shoulders (kg)	0.75	5.95	4.3	0.61
% Weigh losses shoulders	4.41	29.46	23.09	2.77
% Weigh losses ham	0.63	31.95	21.40	3.32

Table 2 shows that there is a significant correlation (p<0.01) between backfat thickness (measured at two different levels) and weigh losses in hams and shoulders during the cutting process (21.40% ± 3.32 for hams and 23.09 ± 2.77% for shoulders). Besides there are significant correlations between ham weigh losses during the cutting process and carcass performance (p<0.05), and between shoulder weigh losses during the cutting process and the slaugther and carcass weight (p<0.01).

Table 2. Pearson correlations between weigh losses during the cutting process and back fat thickness at last thoracic vertebra (BFTult) and fourth lumbar vertebra levels (BFT4) and carcass weigh and performance

	BFT4	BF Tult	Slaughter weigh	Carcass weigh	Carcass performance (%)
Weigh losses of hams during the cutting process	0.86(**)	0.75(**)	-0.04	0.02	0.21(*)
Weigh losses of shoulders during the cutting process	0.29(**)	0.21(**)	0.42(**)	0.51(**)	0.15

*Correlation significance P <0.05; ** Correlation significance P <0.01.

IV – Conclusions

Iberian pigs finished at *montanera* present a positive correlation between the fattening degree of the carcass (measured as back fat thickness) and the weigh losses in hams and shoulders during the cutting process.

Acknowledgements

The authors wish to acknowledge to Turcañada S. L. and Camilo Rios S.L their collaboration

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Physico-chemical and sensorial characteristics evolution of vacuum packaged Iberian dry-cured ham stored at refrigerated temperature

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Abstract. Two groups of five dry-cured hams from Alentejano pig breed submitted to a post-salting cure of 12 months were studied. One of the groups was processed through craft methods at Barrancos (small city of south-east Alentejo region, Portugal) and the other one in a Portuguese meat plant. Equal groups of slices and unitary portions weighing about 250 g were vacuum packed and stored at 7°C. Physico-chemical analysis (pH, a_w, chlorides, total volatile basic nitrogen, thiobarbituric acid index and colour (L*, a*, b*) and sensorial evaluation (colour, off colour, aroma, off aroma, tenderness, succulence, taste, off taste, salt intensity and global evaluation) were carried out before packaging (time 0) and after 2, 5 and 8 months of storage. Statistical analysis consisted in analysis of variance considering the factors "process" (artisanal and industrial), "sample presentation" (portions and slices) and "storage time" (0, 2, 5 and 8 months). Physico-chemical analysis exhibit differences, mainly for "process" factor and sensorial evaluation for "storage time" factor. However, results evidence desirable evolution of the samples during storage time. These study allow to conclude that under refrigeration (7°C) is possible to store vacuum packaged high quality dry-cured ham during, at least, 8 months.

Keywords. Alentejano dry-cured ham – Quality – Storage time – Vacuum packaged.

Évolution des caractéristiques physico-chimiques et sensorielles du jambon de porc Alentejano réfrigéré et conservé sous vide

Résumé. *Deux groupes de cinq jambons de cochon Alentejano avec 12 mois de maturation après salage ont été étudiés. Un des groupes a été traité selon un processus artisanal à Barrancos (sud-est du Portugal) et l'autre groupe a été transformé dans une unité industrielle. Des portions unitaires d'un poids approximatif de 250 g et des tranches de 1 mm d'épaisseur du même poids ont été emballées sous vide et entreposées à 7°C. Les analyses suivantes ont été effectuées : analyses physico-chimiques (pH, aw, chlorures, azote basique volatil total, index d'acide thiobarbiturique, et couleur (L*, a*, b*) et sensorielles (couleur, couleurs étranges, arôme, arôme étrange, tendreté, teneur en jus, saveur, saveurs étranges, intensité de la salaison et évaluation globale) avant l'emballage (temps 0) et après 2, 5 et 8 mois de conservation. Le traitement statistique consistait en une analyse de la variance en considérant les facteurs "processus" (artisanal et industriel), "mode de présentation des échantillons" (portions et tranches) et "temps de conservation" (0, 2, 5 et 8 mois). L'évaluation physico-chimique a montré qu'il y avait des différences et l'analyse sensorielle a montré des différences, en particulier pour le facteur "temps de conservation". Cependant, les résultats ont montré une tendance positive dans les échantillons au cours de la conservation. Cette étude nous permet de déduire qu'il est possible de conserver des jambons de qualité emballés sous vide et maintenus à une température de 7°C pendant au moins 8 mois.*

Mots-clés. *Jambon de porc Alentejano – Qualité – Conservation – Emballage sous vide.*

I – Introduction

Traditional quality products process should be based on scientific and technological support in order to develop poor regions. Recently the production of dry cured ham of Alentejano pig bread increased a lot after a long period of low production due to the reduction of pig production. Due to the high nutritional quality of products that are processed with this specific raw material, the dry cured ham has nowadays a high commercial value that obliges to a high sensorial quality standard. Attributes such as color, fat infiltrated proportion, aroma, textures, and taste are, among other, the most important for consumers and that support the decision of buy the product. Dry cured ham is an expensive food product so the market offers deboned pieces packaged in vacuum conditions. So it´s an interesting and practical objective to study the evolution of deboned packaged ham portions in order to determine their shelf life.

The specific aim of this work is to evaluate the physic-chemical and sensorial characteristics evolution of dry cured ham portions after 12 months of cure, some obtained by artisanal process and others by industrial process, all of them deboned and vacuum packaged, sliced or cute in small portions, and stored at 7ºC along 8 months.

II – Materials and methods

1. Sample preparation

Two groups of five hams of Alentejano pig bread with 12 moths of cure were considered to design this research work. One group was processed in a traditional production, region Barrancos, Alentejo, Portugal. Another group was processed in a modern factory. Sample preparation consists on obtained portions of 250g and slices of 1mm thick all of them packaged at vacuum conditions and stored at 7ºC. Samples were analyzed before being packaged (day 0) and at 2, 5, and 8 moths of storage.

2. Physico-chemical analysis

pH according to NP-3441 (1990), measured with a digital pH meter PTI-9; a_w (Rotronic Hygroscop DT, was measured with a probe WA-40); chlorides according to NP-1845 (1982); total volatile basic nitrogen-ABVT- according to NP-1848 (1987); thiobarbituric acid index-TBA- according to NP-3356 (1987); colour coordinates L*, a*, b* (Minolta CR-210b), were measured on the surfaces of the portions and on the surfaces of the slices.

3. Sensorial evaluation

A panel of 18 panelists evaluated samples made of 1 mm thickness slices. A descriptive quantitative method was used in order to obtain a quantification of different attribute, on structured scale from 0 to 100.

4. Statistical analysis

Statistical analysis consisted in analysis of variance considering the factors "process" (artisanal and industrial), "sample presentation" (portions and slices) and "storage time" (0, 2, 5 and 8 months). For mean comparison LSD test, was used for p<0,05.

III – Results and discussion

There were no significant differences on pH values between samples of artisanal ham and industrial ham (Table1). The highest value of pH was observed at the first day of analysis (time 0). This fact can be justified by the action of microaerophilic lactic acid bacteria, that can be responsible by the lower pH values exhibited by the sliced ham (5.76 for sliced samples and 5.92 for portion samples). The higher development of the lactic acid bacteria can be due to the larger specific surface and so a bigger quantity of substrate available for the bacteria. It was observed that the artisanal ham presented higher values of chlorides due to higher water loose during cure process, without environmental control, also was observed lower a_w values (0.874) than the industrial ham (0.907). During storage period a_w values increased from 0.839 at day 0 until 0.892 after 8 months of storage. This change can be justified by the water released from proteolysis.

Table 1. Means and standard deviation for physic-chemical results

	Process		Sample presentation		Storage time (month)			
	Artisanal	**Industrial**	**Portions**	**Slices**	**0**	**2**	**5**	**8**
pH	5.88 ±0.4	5.82 ±0.2	5.92a ±0.3	5.76b ±0.3	6.08a ±0.08	5.74b ±0.03	5.83bc ±0.04	5.87b ±0.02
a_w	0.874a ±0.004	0.907b ±0.04	0.899 ±0.004	0.900 ±0.004	0.839a ±0.007	0.914c ±0.004	0.894b ±0.003	0.892b ±0.004
Chlorides (%NaCl)	4.9a ±0.1	4.5b ±0.1	4.8 ±0.1	4.6 ±0.1	4.9a ±0.1	4.3b ±0.1	4.8a ±0.1	5.0a ±0.1
ABVT ($mgNH_3$/100g)	230.41a ±14.31	101.34b ±1.97	172.71 ±16.39	156.79 ±12.72	205.67 ±41.15	128.22 ±9.63	165.56 ±15.99	183.95 ±20.27
TBA(mg malonic aldehyde/kg)	0.53a ±0.05	1.40b ±0.27	0.79 ±0.15	1.22 ±0.28	1.33 ±0.49	0.64 ±0.10	0.69 ±0.18	1.47 ±0.43
L*	42.6 ±0.8	41.1 ±1.1	44.2a ±0.9	38.7b ±0.6	39.9 ±1.1	44.9 ±1.1	40.0 ±1.1	41.5 ±1.5
a*	10.8 ±0.5	10.8 ±0.6	9.1a ±0.4	13.1b ±0.4	12.0 ±0.4	12.0 ±0.7	9.7 ±0.7	10.0 ±0.9
b*	5.6a ±0.3	4.2b ±0.4	3.7a ±0.3	6.4b ±0.2	4.2 ±0.6	5.5 ±0.4	4.0 ±0.5	5.4 ±0.6

For each factor and for each physic-chemical variable. different letters represent significantly different means for $p<0.05$.

Probably due to environmental conditions during cure process, more favorable to proteolytic process, were found ABVT values higher in artisanal ham than in industrial ham (230.41 mg NH_3/100g for artisanal ham and 101.34 mg NH_3/100g for industrial ham). Although the higher temperatures during the artisanal cure process, oxidation degree was significantly inferior (0,53 mg of malonic aldehyde) when compared with the values obtained for industrial ham (1,40 mg of malonic aldehyde). Considering that pig nutrition was similar for animals used in both processes and that antioxidant quantity used at the ham preparation were also similar, the differences in TBA values could be caused by a strong contamination by moulds, probably lipolytic, during the cure process and a light exposure of industrial ham at the end of that process. Colour coordinates L* a* b* didn´t show significant differences during storage time, and sliced ham always present different values when compared with portions, for all the coordinates, L* with 44.2 for portions and 38.7 for slices, a* with 13.1 for slices and 9.1 for portions, and b* with 6.4 for slices and 3.7 for portions. So the sliced ham seems to change more than portions.

About the results of sensorial evaluation (Table 2) it could be observed that values obtained for colour after 5 and 8 months of storage (66 and 67 points respectively) were significantly higher than those at 0 and 2 months (56 and 54 months respectively). Aroma attribute was classified with significantly superior values for artisanal ham (58 points) than industrial ham (53 points) probably due to intensive proteolytic and lipolytic phenomena occurred in artisanal ham. Sliced ham had better classification than portions for the attributes tenderness and taste. Taste was better after 6 or 8 months of storage (62 and 60 points respectively) than at time 0 and 2 months of storage (52 point for both). Global evaluation was almost similar for artisanal ham and industrial ham (55 and 57 points respectively). However there were noticeable differences between slices and portions for global evaluation with higher values for the first samples, slices. On the other hand time storage improved sensorial global evaluation of ham with (58 points after 8 storage months and 50 points for time 0). Panel noticed strange colours, strange aromas and strange taste but always at low levels. Storage time never caused the increasing of those undesirable attributes. Aroma and strange taste can be related to the presence of acetaldehyde and pentane (results not shown in this work).

Table 2. Means and standard deviation for sensorial results using a scale from 0 to 100

	Process		Sample presentation		Storage time (month)			
	Artisanal	**Industrial**	**Portions**	**Slices**	**0**	**2**	**5**	**8**
Colour	60±2	63±1	62±1	61±2	56a±2	54a±2	66b±1	67b±2
Strange colours	5±1	6±1	6±1	5±1	7ab±1	8b±1	5ab±1	3a±1
Aroma	58a±1	53b±1	53a±1	60b±1	48a±5	50a±3	62b±2	59b±1
Strange aromas	5±1	3±0	4±1	3±1	7a±2	4ab±0	3b±0	3b±1
Tenderness	58±1	58±1	55a±1	62b±1	55ab±0	53b±1	62c±2	61bc±3
Suculence	52±1	49±1	48a±1	52b±1	43a±1	45a±1	57b±2	51c±1
Taste	58±1	56±1	56±1	59±1	52a±1	52a±2	62b±2	60b±2
Strange taste	8±1	4±1	7±1	5±1	10±2	6±1	5±2	6±3
Salt intensity	50±1	43±1	47±1	45±1	43ab±4	42b±2	48ac±2	50c±2
Global evaluation	55±1	57±1	54a±1	59b±1	50a±1	52a±1	61b±1	58b±3

For each factor and for each physic-chemical variable. Different letters represent significantly different means for p<0.05.

IV – Conclusions

Physico-chemical parameters were different for artisanal and industrial hams. The different sample presentation didn´t influence a lot the physic-chemical parameters and, considering a practical point of view, the changes observed along the storage time weren't also very expressive.

The sensorial panel didn't notice important differences between the artisanal and industrial hams, neither between sliced ham and portions, however panelists preferred samples stored for a long time. Colour, aroma, tenderness, succulence, flavor became better with the permanence inside the package.

To storage portions and slices of dry cured ham of Alentejano pig bread in vacuum conditions at cold temperature (7ºC) is possible for at least 8 months, without noticeable changes in desirable attributes.

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Effect of fermentation temperature and nitrite nitrate on properties of dry fermented sausage

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Abstract. Sausages, produced with/without nitrite and nitrate, were fermented at 21°C (RH: 65-74%) or 8°C (RH: 75-85%). All sausages were dried until a final weight loss of 44% and analyzed for physicochemical and microbiological data. The removal of NO_2^-/NO_3^- caused an increase in lipid oxidation, although the decrease in the fermentation temperature proved to be effective in controlling the lipid oxidation of ripened nitrite-free sausages. The effect of nitrite on the formation of red and bright colour was evident since the early stages of processing, while its removal resulted in the formation of a less red, less stable and lighter colour, in spite of fermentation temperature. The texture of ripened sausage did not show significant differences among groups. The elimination of NO_2^-/NO_3^- had no effect on the typical microbiota (LAB and SNCP), while differences were observed for *B. thermosphacta*, enterococci and enterobacteria.

Keywords. Colour – Dry fermented sausage – Fermentation temperature – Microbiota – Nitrite – pH.

Effet de la température de fermentation et de l'élimination des nitrites et nitrates sur les paramètres du saucisson sec

***Résumé**. Des saucissons produits avec et sans nitrates et nitrites, ont été fermentés à 21°C (RH 69-74%) et à 8°C (RH 75–85%). Les saucissons ont été maturés jusqu'à une perte de poids de 44%. Les analyses physico-chimiques et microbiologiques étaient effectuées au cours de la maturation. L'élimination de NO_2^-/NO_3^- augmente l'oxydation des lipides mais la réduction des températures de fermentation contrôle l'oxydation des saucissons qui ont été produits sans nitrates. L'importance des nitrites dans la formation de la couleur rouge vif est évidente dès le début de leur production; leur élimination, indépendamment des températures de fermentation, détermine la formation d'une couleur rouge moins intense, moins stable et plus claire. Les paramètres de texture du salami maturé n'ont pas mis en évidence de différences significatives entre les groupes. L'élimination de NO_2^-/NO_3 n'affectait pas le microbiote typique (LAB et SNCP) tandis que des différences ont été observées à propos de* B. thermosphacta*, entérocoques et* Enterobacteriaceae*.*

***Mots-clés.** Couleur – Saucisson sec – Températures de fermentation – Microbiote – Nitrate – pH.*

I – Introduction

The use of nitrite in sausage production is important for its antibacterial, colour-forming, antioxidant and flavouring properties (Shahiti and Pegg, 1992). Despite these properties, there are problems with regard to toxicity of nitrite and nitroso-derivatives for human health (Cassens, 1997). The processing of the sausage is based on a drip and drying phases which reduce water activity; between these two phases there is a fermentation under controlled temperature and relative humidity, during which the growth of lactic acid bacteria, results in a pH decrease, thus the development of texture and colour and the control of spoilage bacteria and pathogens (Hugas and Monfort, 1997; Lucke, 1998). The aim of this research was to determine the effects of fermentation temperature and nitrite nitrate on the physicochemical and microbiological characteristics of dry fermented sausages.

II – Materials and methods

1. Sausage formulation and processing

Three batches of dry fermented sausages, two without NO_2^-/NO_3^- (batches N-) and one with 80 ppm $NaNO_2$ and 120 ppm KNO_3 (batch N+) were produced from a common meat batter (approximately 75% lean pork meat and 25% pork fat). The pork lean and fat were ground through a 7 mm diameter mincing plate and mixed with salt (2.4%), sucrose (0.25%), sodium ascorbate (0.06%), white wine, whole black pepper, ground white pepper and garlic. A commercial starter culture containing *Lactobacillus curvatus, Staphylococcus carnosus and Kocuria varians* was added. Batters were stuffed into natural casing (80-85 mm diameter), and the final weight for each sausage was 1 kg. A mould starter, SK10 *Penicillium nalgiovensis*, was applied on the surface of sausages. For the batches N- two different processes (P) were applied: "P1" consisting of 3 d at 21±1ºC /65-74% relative humidity (RH) followed by a drying at 17±2ºC/70-80% RH and ripening at 13±1°C/80-88 % RH; "P2" consisting of 13 d at 8±1ºC/75-85% RH. Afterwards the temperature was increased to 16±1°C and 80-88% RH for 7 d and finally 18±1°C and 80-88% RH until the end of the processing. The sausages produced with NO_2^-/NO_3^- (batch N+), were processed with P1 only. All sausages were processed until a final weight loss of 44% was reached. From each batch, samples of the sausage batter (100 g) were collected at days 0 (prior to stuffing) and three sausages were taken at 6, 13 and 29 d. At the end of the process 5 sausages for each batch were taken for analysis.

2. Analysis

Water activity (a_w) was measured at 25ºC by means of Aqualab® equipment (Model Series 3TE, Decagon Devices Inc). *pH* was measured using a pH meter WTW (model 330) inserting the electrode (Hamilton) directly into the sausage. *Lipid oxidation (TBArs)* was determined using the 2-thiobarbituric acid method (TBA-test) described by Witte *et al*. (1970) using trichloroacetic acid 5% as solvent. *Textural Profile Analysis* (Bourne, 1978) was performed *(*Instron Texture Machine mod. 5565) using the central core of two slices of each sample. Each probe (15 mm height and 25 mm diameter) was compressed twice to 50% of original height. The following texture parameters were calculated: hardness (peak force during the 1st compression cycle), and cohesiveness (ratio of the positive force area during the 2nd compression to that during the 1st compression). *Colour* (Minolta CR-508d; illuminant D65), was measured by the CIE L*a*b* system and the results were expressed as lightness (L*), redness (a*) and yellowness (b*). The sausages were cut into sections 2 cm thick and colour measurements were taken immediately after cutting, and after 24 hr of display in air a 4°C. Sausages were subjected to the following microbiological analyses: *Enterobacteriaceae* (ISO 7402), Gram-negative bacteria (G-) (violet red bile glucose agar 30°C/48 hr), Lactic acid bacteria (LAB) (ISO 15214), Micrococci and staphylococci (Mannitol Salt Agar, 30°C/72 hr) and enterococci (Slanetz Bartley agar 37°C/ 4hr and 44°C/44 hr).

The chemical-physical and microbiological data were shown as mean values. One-way analysis of variance, (ANOVA, SPSS vr.11.5.0) was run to detect differences among batches at end of ripening (Bonferroni test).

III – Results and discussion

Figure 1 shows the weight losses of sausages throughout the ripening processes: a reduction approximately 44% (w/w) was reached in about 61 d for sausages processed with P1 and 67 d for sausages processed with P2, meaning that the slow ripening conditions of P2 were controlled to minimize differences in drying from a traditional process. During the processing a_w dropped from the initial value of 0.97 to the final average value of 0.86, without significant differences between the three batches.

All sausages showed at first a pH decrease and a final rise (Fig. 2a). The rate of pH decrease was affected by fermentation temperatures: the higher temperature in P1, favouring formation of organic acids, caused a faster pH decrease than in P2 (6 d vs 13 d). At the end of fermentation and at the end of ripening the pH values of batch N+ were lower ($p<0.005$) than in batches N-.

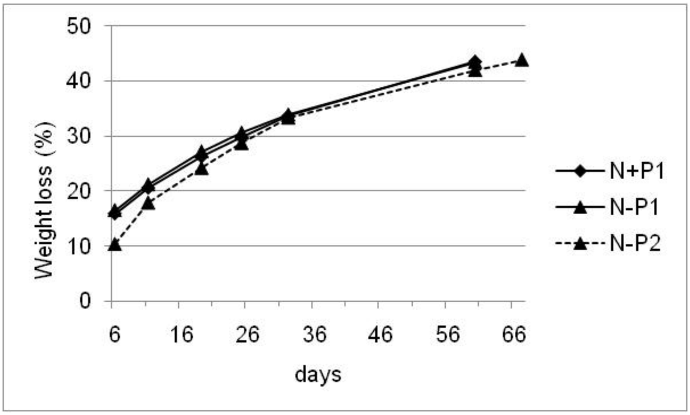

Fig. 1. Mean values of weight loss in the different batches during the ripening of sausages (N-P1: no NO_2^-/NO_3^- added sausage, process P1; N-P2: no NO_2^-/NO_3^- added sausage, process P2; N+P2: NO_2^-/NO_3^- added sausage, process P2).

For all batches TBArs values showed an increase followed by a reduction(Fig. 2b). The lipid oxidation was more rapidly increased in N-P1 than in N-P2 or in N+P1 batches but, at 13 d the highest TBArs values were observed in both N- batches. At end of ripening process, ANOVA revealed that oxidation of N-P1 sausages was higher ($p=0.017$) than N+P1 and N-P2.

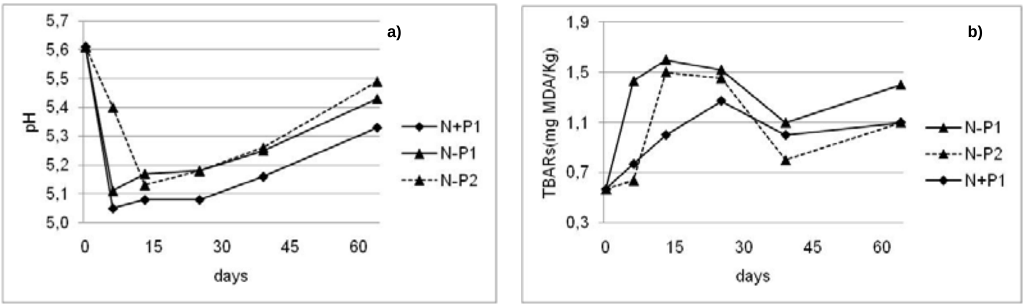

Fig. 2. Mean values of pH (a) and TBArs (b) in the different batches during ripening.

The effect of nitrite on the formation of a red and bright colour was evident since the early stages of processing (data not show), and its removal resulted, at end of ripening, in the formation of a less red, more yellow and lighter colour (Fig. 3). The storage (24 hr at 4°C) of sliced sausages determined an increase of differences in color index among batches N+ and N- despite processing condition.

pH and moisture are major factors affecting texture properties and the increase of hardness (maximum force required to compress the sample) and cohesiveness (strength of the internal bonds making up the body of the sample) observed during ripening. The observed differences of hardness and cohesiveness (Fig. 4) among the batches during the first phases of ripening could be explained by the differences in the rate of pH decrease and in the weight loss. At end of ripening process, the texture parameters of sausages did not show significant differences among batches.

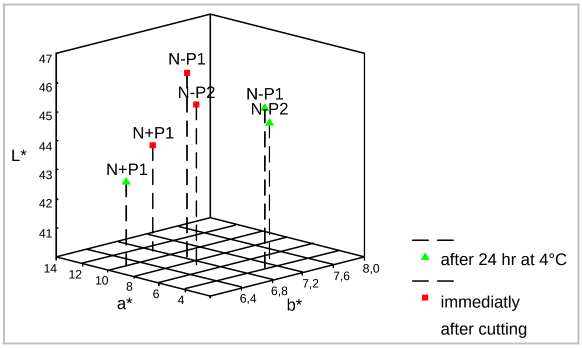

Fig. 3. Colour parameters, after cutting and after 24 hr at 4°C, of the ripened sausage.

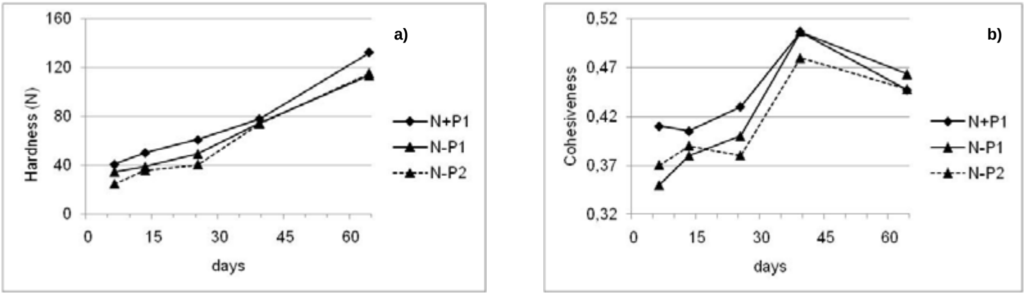

Fig. 4. Mean values of hardness and cohesiveness in the different batches.

Figure 5 shows the fate of typical and spoilage microorganisms during the processing of different batches of sausages. In P1 the elimination of NO_2^-/NO_3^- had no effect on the typical microbiota: LAB increased during the first 10 d, reaching values in the order of 10^9 cfu/g, staphylococci not coagulase positive (SNCP), introduced with the starter at level of 10^6 to 10^7 cfu/g, remained the same for the entire process. Differences were observed for the behaviour of G-, *B. thermosphacta* and enterococci. Trends of G-, in the batch N+P1 revealed, in the first 10 d, a drastic and progressive reduction to reach at the end of process values less than 10 cfu/g; *B. thermosphacta* showed a similar pattern. In batch N-P1 a change was observed in the load of G- that after 30 d, were still around 10^3 cfu/g and only at the end of maturing showed values <10 cfu/g. The behaviour of *B. thermosphacta* during the first 30 d is almost similar to that of the batch N-P1 but, at the end of processing, a great variability (10-10^4 cfu/g) among the values, was detected in the different samples. In the N-P2 batch the use of temperature as low as 8°C during the fermentation allowed more control over the evolution of the spoilage bacteria. G- bacteria decreased steadily reaching, after maturing, values <10cfu/g; enterococci remained at the same level until the end of processing. *B. thermosphacta*, after an initial reduction of about 2 log, showed a sharp increase and, at the end, the level was higher than initial one.

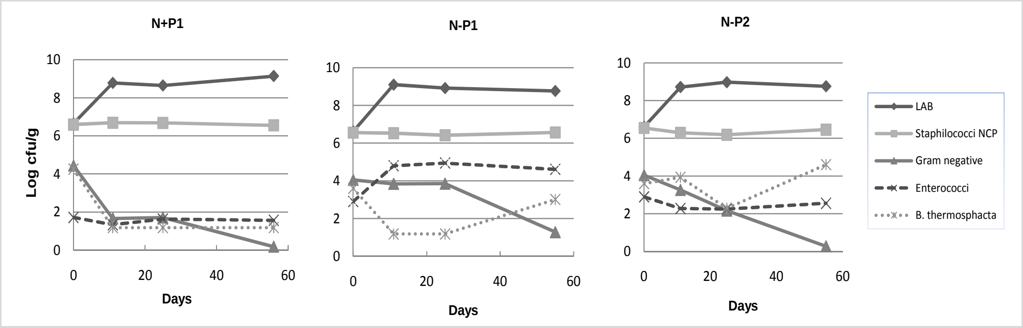

Fig. 5. Performance of the main microbial groups during ripening of sausage.

IV – Conclusions

Under the conditions of the present study, the production of dry cured sausages without nitrate and nitrite resulted in a not very stable colour and in an uncontrolled lipid oxidation specially at higher temperature. No effect on the typical microbiota was detected whereas spoilage bacteria growth wasn't controlled. The slower rate of acidification at low temperature (8°C) of fermentation did not significantly affect the weight loss texture parameters and it allowed more control of the evolution of Gram negative and enterococci.

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Volatile hydrocarbon profile of Iberian dry-cured hams. A possible tool for authentication of hams according to the fattening diet

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Abstract. The aims of this work were to carry out a comprehensive study of the volatile hydrocarbons of 34 Iberian dry-cured hams and to evaluate the efficiency of these compounds for discriminating hams according to the fattening system: "Montanera" (B) and "Cebo" (C). The samples of hams were obtained by mincing the semimembranosus and semitendinosus muscles from slices of dry-cured ham. The analyses were carried out by gas chromatography-mass spectrometry with a polar capillary column and after a previous extraction by Purge and Trap method. Forty-three volatile hydrocarbons were identified, 26 of them for the first time in Iberian dry-cured ham. Only five compounds showed significant differences between the two types of hams. Among the 33 volatile hydrocarbons, 22 of them allowed a complete discrimination of the two groups of hams according the fattening system.

Keywords. Iberian pig – Slice ham – Volatile hydrocarbons – GC-MS.

Profil en hydrocarbures volatils chez des jambons ibériques. Un instrument éventuel pour vérifier l'authentification des jambons en fonction de la diète

Résumé. *L'objectif de ce travail est de mener une étude concernant les hydrocarbures volatils sur 34 jambons ibériques et d'évaluer l'efficience de ces composés en vue de différencier les jambons en fonction du système d'alimentation: "Montanera" (B) et "Cebo" (C). Les échantillons de jambons ont été obtenus sur des tranches de jambon, plus précisément des mélanges des muscles* Semimembranosus *et* Semitendinosus. *L'analyse a été réalisée par GC-MS avec une colonne polaire après extraction par une méthode de "purge and trap". Quarante-trois hydrocarbures volatils ont été identifiés, parmi lesquels 26 l'ont été pour la première fois chez des jambons ibériques séchés. Seuls cinq composés ont montré des différences significatives entre les deux types de jambons. Sur les 33 hydrocarbures volatils, 22 ont permis que les deux groupes de jambons étudiés se différencient complètement concernant leur système d'alimentation.*

Mots-clés. *Porc Ibérique – Tranche de jambon – Hydrocarbures volatils – GC-MS.*

I – Introduction

Several authors (López *et al*., 1992) have identified a large number of volatile compounds such as aldehydes, ketones, aliphatic hydrocarbons, aromatic hydrocarbons, alcohols, carboxylic acids, esters and lactones in the Iberian dry-cured hams. It has been postulated that these compounds arise from numerous chemical or enzymatic reactions such as lipolysis, chemical or enzymatic oxidation, proteolysis, Strecker degradation and Maillard reactions (Toldrá *et al*., 2000). Most of these studies on volatile compounds have been carried out with the aim of characterizing them or describe their contribution to the flavour of dry-cured hams. Only an attempt to explore the utility of these compounds as classifying factor for the fattening diet has been carried out, but in loins not in hams. The aims of this work were to carry out an exhaustive study of the volatile compound fraction of 34 Iberian ham samples and to explore the utility of these compounds, mainly short chain hydrocarbons, as discriminating factors for the fattening diet system.

II – Experimental design

1. Ham samples

A total of 34 samples of dry-cured hams from castrated male 14-month-old pure Iberian pigs and processed in an industry for 24 months were used: 23 corresponding to animals of "Montanera" and 12 corresponding to animals of "Cebo". Slices were cut parallel to the femur and to different depths form each ham. Each slice contained semimembranosus and semitendinosus muscles were trimmer by removing the adipose tissue.

2. Methods

The volatile hydrocarbons were analysed by the dynamic headspace technique and adsorbed on a Tenax trap, using a Purge and Trap Concentrator apparatus Tekmar velocity XPT (Thousand Oaks, CA, USA), based on the method described by Sabio *et al*. (1998), one of the most useful analytical methods to determine volatile compounds. After, the volatile compounds were desorbed by heating, the Tenax trap at 225ºC for 1 min, and sent throw of transfer line (dept at 150 ºC) into the chromatograph injector. The GC-ion-trap-MS analyses were performed using a Varian 3800 gas chromatograph coupled to a Saturno 2000 ion trap mass spectrometer (Varian, Palo Alto, CA, USA).The identification and quantification of the volatile hydrocarbons was done comparing the spectra with those from NIST (National Institute of Standars and Technology) and WILEY libraries and verified by standards.

III – Results and discussion

1. Volatile hydrocarbons profile of ham

A total of 43 volatile hydrocarbons have been identified by GC-MS (Fig. 1). The different hydrocarbons identified in the volatile fraction from "Montanera" and "Cebo" samples are shown in Table 2. Together with mean values, standard deviation (S.D.), maximum and minimum values.

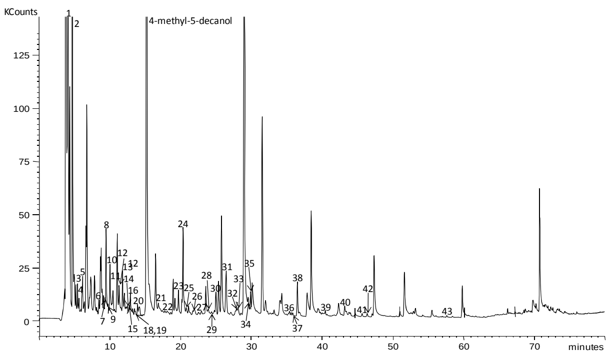

Fig. 1. Chromatograms of the volatile compounds profile of Iberian ham slice samples.

In the group of linear and branched hydrocarbons, 2,4-dimethyl-heptane, 2-octene, 2,2,5,5-tetramethyl-hexane, 2,25-trimethyl-hexane, 2,3,5,8-tetramethyl-decane, 4-methyl-1-decene, 2,4,6-trimethyl-heptane, diisoamilene, 7-methyl-pentadecene, 2,2,3-trimethyl-nonane, 5-(1-

methyl-propyl)-nonane and 3-methyl-5-undecene are observed for the first time in the volatile fraction of Iberian ham.

Table 1. Volatile hydrocarbons profile of Iberian dry-cured ham samples

No.	Volatile hydrocarbons	*"Montanera"*				*"Cebo"*			
		mean	SD	max	min	mean	SD	max	min
	n-alkanes								
5	Nonane	0.36	0.28	0.91	0.00	0.25	0.15	0.45	0.05
8	Dodecane[a]	2.65	3.42	10.10	0.00	0.40	0.28	0.89	0.00
	Branched alkanes								
1	3-methyl-hexane	4.64	4.90	23.46	0.56	4.17	1.14	5.93	2.37
2	2,4-dimethyl-heptane[a]	4.65	3.11	11.85	0.89	7.04	3.53	14.28	0.86
7	2,2,5,5-tetramethyl-hexane[b]	0.12	0.28	1.12	0.00	0.48	0.42	1.15	0.00
9	2,2,5-trimethyl-hexane	0.84	1.14	3.20	0.00	0.21	0.25	0.70	0.00
10	2,3,5,8-tetramethyl-decane	1.78	1.74	5.38	0.05	0.98	0.94	2.67	0.00
14	2,4,6-trimethyl-heptane[a]	0.60	0.54	1.77	0.00	0.23	0.19	0.54	0.02
16	7-methyl-pentadecane	0.53	0.59	1.83	0.00	0.38	0.47	1.28	0.00
17	2,2,3-trimethyl-nonane	0.31	0.41	1.51	0.00	0.25	0.35	0.98	0.00
18	5-(1-methyl-propyl)-nonane	0.15	0.17	0.52	0.00	0.10	0.14	0.39	0.00
21	2,6-dimethyl-undecane	0.11	0.12	0.41	0.00	0.05	0.08	0.24	0.00
	n-alkenes								
4	2-octene[a]	0.21	0.14	0.49	0.00	0.11	0.08	0.25	0.01
	Branched alkenes								
12	4-methyl-1-decene	1.64	1.58	4.83	0.06	0.90	0.66	2.30	0.10
26	3-methyl-5-undecene	0.12	0.18	0.58	0.00	0.14	0.19	0.68	0.00
27	4-methyl-1-undecene	0.18	0.21	0.93	0.00	0.26	0.30	1.08	0.03
	Cyclic								
3	1,2-diethyl-cyclobutane	0.17	0.10	0.41	0.00	0.35	0.56	2.06	0.05
6	Butyl cyclopentane	0.16	0.20	0.93	0.00	0.15	0.23	0.87	0.00
20	Heptyl-cyclohexane	0.12	0.26	1.16	0.00	0.29	0.35	0.97	0.00
22	1-ethyl-1-methyl-cyclohexane	0.08	0.16	0.73	0.00	0.10	0.14	0.46	0.00
29	Octyl-cyclohexane	0.06	0.15	0.64	0.00	0.18	0.31	0.99	0.00
37	2-ethenyl-cyclohexane	0.03	0.07	0.29	0.00	0.02	0.02	0.05	0.00
39	Butenyl cyclohexene	0.03	0.04	0.17	0.00	0.02	0.03	0.09	0.00
43	*cis*-1,2,3,4-tetramethyl-cyclopentane	0.14	0.30	1.25	0.00	0.07	0.12	0.31	0.00
	Terpenic								
31	Limonene	0.49	1.14	5.41	0.00	1.12	1.39	4.09	0.03
11	4-carene	0.88	0.95	2.59	0.00	0.36	0.27	1.00	0.06
19	Germacrane B[a]	0.35	0.30	1.05	0.00	0.11	0.13	0.31	0.00
	Aromatic								
13	Methyl-benzene	2.10	3.07	13.42	0.34	0.43	0.23	0.96	0.14
15	Diisoamilene	0.34	0.41	1.67	0.00	0.24	0.22	0.67	0.00
23	p-xylene	0.30	0.20	0.79	0.00	0.30	0.25	0.76	0.05
24	m-xylene	0.61	0.72	3.15	0.00	0.37	0.32	1.15	0.00
25	Decahydro-*cis*-naphtalene	0.31	0.50	2.08	0.00	0.04	0.06	0.18	0.00
28	2-methyl-decahydronaphtalene	0.10	0.19	0.70	0.00	0.08	0.23	0.81	0.00
30	o-xylene	0.14	0.16	0.60	0.00	0.33	0.38	1.16	0.00
32	Propyl-benzene	0.11	0.15	0.73	0.00	0.09	0.14	0.40	0.00

Table 1. Volatile hydrocarbons profile of Iberian dry-cured ham samples (cont.).

No.	Volatile hydrocarbons	"Montanera"				"Cebo"			
		mean	SD	max	min	mean	SD	max	min
33	Decahydro-*trans*-naphtalene	0.05	0.08	0.24	0.00	0.11	0.15	0.37	0.00
34	1,3,5-trimethyl-benzene	0.03	0.07	0.30	0.00	0.09	0.14	0.43	0.00
35	1-ethyl-4-methyl-benzene	0.05	0.13	0.61	0.00	0.03	0.09	0.30	0.00
36	1-methyl-3-(1-methyl-ethyl)-benzene	0.04	0.09	0.45	0.00	0.07	0.08	0.23	0.00
38	1,2,4-trimethyl-benzene	0.15	0.20	1.00	0.00	0.12	0.08	0.30	0.05
40	1,2,3-trimethyl-benzene	0.11	0.31	1.50	0.00	0.02	0.02	0.07	0.00
41	4-ethyl-1,2-dimethyl-benzene	0.02	0.04	0.16	0.00	0.00	0.01	0.04	0.00
42	2-ethyl-1,3-dimethyl-benzene	0.01	0.02	0.07	0.00	0.00	0.01	0.04	0.00

[a] For $p < 0.05$; [b] For $p < 0.01$.

All cyclic hydrocarbons except limonene: 1,2-diethyl-cyclobutane, butyl-cyclopentane, germacrane B, heptyl-cyclohexane, 1-ethyl-1-methyl-cyclohexane, octyl-cyclohexane, 2-ethenyl-cyclohexane, butenyl-cyclohexane and *cis*-1,2,3,4-tetramethyl-cyclopentane compounds have been identified for the first time in the present work. On the other hand, in the present study, we have identified the 4-carene, which has not been described previously.

Most of the aromatic hydrocarbons have been previously described by other authors, however, decahydro-*cis*-naphtalene, decahydro-*trans*-naphtalene, 2-methyl-decahydronaphtalene and 2-ethyl-1,3-dimethyl-benzene have been described for the fist time in this work. Besides, (1-methyl-propyl)-benzene, 1-propenyl-benzene and 1-methyl-4-(1-methyl-ethenyl)-benzene compounds were detected and they have not been described at the literature previously.

A PCA was performed (Fig. 2) where it shows a fair separation between "Montanera" (B) and "Cebo" (C) samples. To achieve a better separation of the groups according to fattening diets a linear discriminant analysis (LDA) was carried out. Fig. 3 shows the case discrimination, grouped by fattening diet, according to the first canonical variable or square roots obtained from the classification function. A complete separation between the two groups can be observed.

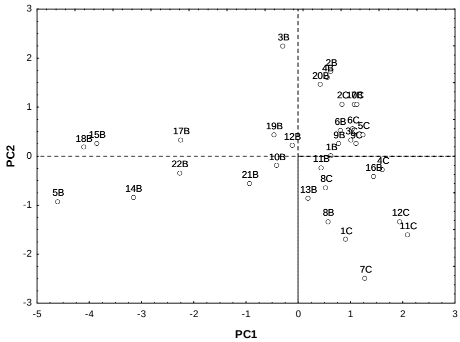

Fig. 2. Principal Component Analysis (PCA).

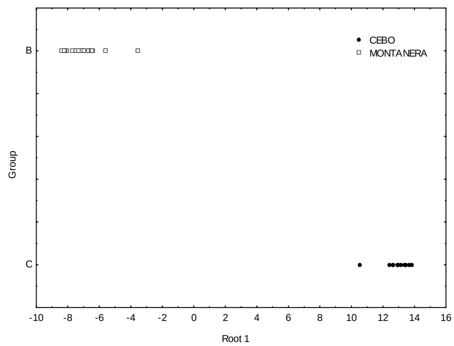

Fig. 3. Linear Discriminant Analysis (LNA).

Acknowledgments

The authors are grateful to the Designation o Origin "Los Pedroches" for the collaboration and given help. This study was supported by projects PET 2007-0015 and P08-AGR-03498.

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Session 6
Quality assurance and traceability

Quality of dry cured ham: Methods for authentication of geographical origin, rearing system and technology

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Abstract. Traceability and authentication of meat and meat products are major concern for consumers, producers and retailers. In Europe, several areas produce high quality dry cured hams under Protected Designation Origin. Efficient and objective methods must be developed to assess origin of dry cured hams. This paper is focused on methods to assess geographical origin, rearing systems and processing conditions. Methods to assess geographical origin are based on multi-analyses of both stable isotopes ($^{18}O/^{16}O$, $^{2}H/^{1}H$, $^{15}N/^{14}N$, $^{13}C/^{12}C$, $^{34}S/^{32}S$) and trace minerals (Se, Fe, Sr, Cu, Zn…). Genotypes of pigs can be check by biotechnological methods based on DNA analyses (RFLP, microsatellites). Methods to trace feeding system are based on stable isotope measurement or on quantification of organic components (hydrocarbons, poly phenols, fatty acids, vitamins) in muscles and adipose tissues. Up to now, very few methods have been focused on the authentication of specific processing conditions. Up to now, most of the methods remain at the stage of potential tools and needs to be performed on large numbers of samples to be applied with certainty in routine controls. A proper authentication of dry cured hams requires a combination of methods and multivariate statistical analyses. One of the main challenges for the future is to build wide opened data bases easy to use, aggregating data available on dry cured hams.

Keywords. Authentication – Dry cured ham – Geographical origin – Analytical method.

Qualité du jambon sec : Méthodes pour l'authentification de l'origine géographique, du système d'élevage et de la technologie

Résumé. *La traçabilité et l'authentification des viandes et des produits carnés est un sujet majeur de préoccupation pour les consommateurs, producteurs et détaillants. En Europe, plusieurs régions produisent des jambons secs de grande qualité sous appellation contrôlée. Des méthodes efficaces et objectives doivent être développées pour vérifier l'origine des jambons secs. Cet article est focalisé sur les méthodes destinées à vérifier l'origine géographique, les conditions d'élevage et de transformation. Les méthodes consacrées à l'identification de l'origine géographique reposent sur une analyse multiple d'isotopes stables (*$^{18}O/^{16}O$, $^{2}H/^{1}H$, $^{15}N/^{14}N$, $^{13}C/^{12}C$, $^{34}S/^{32}S$*) et de minéraux (Se, Fe, Sr, Cu, Zn…). Le génotype des porcs est vérifié par des méthodes de biotechnologie basées sur l'analyse de l'ADN (RFLP, microsatellites). Les méthodes pour tracer les modalités d'élevage reposent sur la mesure des isotopes stables ou la quantification des composés organiques (hydrocarbures, polyphénols, acides gras, vitamines) dans les muscles et les tissus adipeux. À ce jour, peu de méthodes ont été consacrées à la traçabilité des conditions de transformation. Présentement, la majorité des méthodes restent au stade d'outils potentiels et nécessitent d'être validées sur de grands nombres d'échantillons pour être utilisées en routine. Une identification correcte des jambons secs requiert une combinaison de méthodes et d'analyses statistiques multivariantes. Un des enjeux majeurs pour le futur est de construire des bases de données largement ouvertes agrégeant les données disponibles sur les jambons secs de manière intelligible.*

Mots-clés. *Authentification – Jambons secs – Origine géographique – Méthodes analytiques.*

I – Introduction

Traceability and authentication of meat and meat products are a major concern not only for consumers but also for producers and retailers (Karoui *et al*., 2007). Thus, the globalization of food market allows a worldwide transportation of both raw material and processed meat that make easy malpractices and substitution of high quality raw material and products by lower quality ones. The international exchanges are also involved in the outbreak of worldwide diseases related to meat consumption (BSE) or animal farming (avian influenza, Foot and Mouth disease).

Consequently, for the last 20 years, the European Union has been reinforcing the policy demanding to all the food companies to develop efficient methods to trace food from farm to fork and also from fork to farm (Schwagele, 2005). In parallel, The EU has promulgate regulation "On the protection of geographical indications and designations of origin for agricultural products and foodstuffs" (Council regulation, 2081/92) for the protection of food names based on geographical origin (Protected Designation of Origin – PDO, Protected Geographical Indication – PGI) or on traditional recipe (Certificate of Specific Character – CSC).

The main goals of these regulations are to:

(i) Restore the confidence of consumers in meat and meat products giving right information on products to guide their choices;

(ii) Develop a comprehensive and integrated food safety policy and prevent food crisis;

(iii) Ensure fair trade and right prices for high quality products (Schwagele, 2005).

If regulations are required to prevent voluntary and involuntary mislabeling of food and food products, they are not sufficient. Effective analytical methods are required to deliver objective proofs that the label products were produced to meet the requirements described in specifications of PDO, PGI or CSC.

Numerous methods have been proposed to authenticate geographical origin or specific process of various foods (Perez *et al*., 2007; Muller and Steinhart, 2007; Luykx *et al*., 2008). They can be divided into two groups (Perez *et al*., 2007). The first one includes methods based on fast and non destructive methods using spectroscopic characteristics of products. These methods provide fingerprints of foods and often require complex data analyses. They remain very difficult to relate to the specifications of products. The second one includes methods based on chemical analyses of raw material or end-product. Often more tedious these methods can be easily interpreted because they are based on an extensive scientific knowledge on the relationship between environment, production system and processing conditions on quality traits of end-products. Up to now, more than thousand papers have been published on the main labeled dry cured hams produced in Europe. They describe the relationships between traits of raw material or dry cured products and genotypes (Gandemer, 2002, Tejeda *et al*., 2002), rearing systems of pigs (Lebret *et al*., 1996, Coutron-Gambotti *et al*., 1999, Andres *et al*., 2001) and processing conditions (Gandemer, 2002, Andrès *et al*, 2004). The main requirements written in specifications of labeled dry cured hams are supposed to be involved in the typical traits of dry cured hams of each area of production (Flores, 1997, Gandemer, 2009). Thus, rearing and feeding systems including breed, age at slaughter and consumption of local feeds during fattening largely affect raw material chemical composition (Gandemer, 2002*;* Ruiz et Lopez-Bote, 2002). The conditions of processing affect the chemical, physical and organoleptic traits of dry cured hams through a set of complex reactions of lipolysis, oxidation and proteolysis, kinetics of those largely determined by temperature and length of the different steps of the process (Gandemer, 2002*;* Toldra et Navarro, 2002, Toldra, 2006). In contrast, few papers have been published on composition in micronutrients such as minerals or in the ratio of stable isotope in muscle or adipose tissues from pigs and on the environment where they are reared.

Up to now, most of the available methods have been developed to assess the origin of plant foods (Olive oils, wines…) (Kelly *et al*., 2005*;* Gonzalvez *et al*., 2009). These for meat and meat products authentication are less numerous (Ballin, 2010). Most of them are not yet effective tools but just potential ones because in many cases they have been established on a too small set of animals or dry cured hams often of well known origins.

II – Geographical origin

The ratios of stable isotopes of components that constitute all the biological tissues such as muscles and adipose tissues depend on many factors but some of them are strongly related to geographical origin ($^{18}O/^{16}O$, $^{2}H/^{1}H$, $^{15}N/^{14}N$, $^{13}C/^{12}C$, $^{34}S/^{32}S$)(Karoui and de Baerdemaeker, 2007). Thus, $^{18}O/^{16}O$ and $^{2}H/^{1}H$ ratios in water depend on parameters such as the altitude, the distance to ocean and the climate. $^{15}N/^{14}N$, $^{13}C/^{12}C$, $^{34}S/^{32}S$ ratios depend on organic matter in soil and fertilizers. The amount and composition of trace elements in soil (Se, Fe, Sr, Cu, Zn…) are strongly related to the geological underground or specific pollutions from human activities (mining, accident). These elements are incorporated into animal tissues through food chain (Franke *et al*., 2005).

Measurement of $^{18}O/^{16}O$ and $^{2}H/^{1}H$ in tissue water is an interesting tool for geographical origin assessment because these ratios are strongly correlated to these in drinking water (Karoui and de Baerdemaeker, 2007*;* Heaton *et al*., 2008). Compared to the ratio of these stable isotopes in ocean water, these ratios are lower in altitude or far from the ocean because stable isotopes are discriminated through the successive cycles of evaporation, condensation and precipitation. The ratios of stable isotopes in water are very good indicators of meat origin because they are only slightly affected by feeding systems and main part of the body water come from drinking water. These methods were used with success to discriminate milk products (Heaton *et al*., 2008*;* Karoui and De Baerdemacher, 2007) and beef meat from different continents (Boner and Förstel, 2004; Schmidt *et al*., 2005*;* Horacek and Min). No data is available on pig meat. But discriminating pig meat from European areas of production of high quality dry cured hams could be very difficult because the main areas of production are close to each other, close to the ocean and in mid-mountains. So the isotope ratio in water could be too close to discriminate geographical origin of meats.

In some studies, $^{15}N/^{14}N$ and $^{13}C/^{12}C$ ratio in proteins or lipids of meat were used to discriminate beef and lamb meat according to their geographical origin (Karoui and de Baerdemaeker, 2007; Piasentier *et al*., 2003). The principle is based on the fact that plants from tropical countries are mainly C4 plants while those from temperate countries are mainly C3 plants. C3 plants discriminate more ^{13}C and exhibit a lower $^{13}C/^{12}C$ than C4 plants. Consequently, animals eating more C3 plants have a lower $^{13}C/^{12}C$ in their tissues. However, the quantification of $^{15}N/^{14}N$ and $^{13}C/^{12}C$ suffers of serious drawbacks related to feeding systems (see next part) or to agricultural practice such as fertilizers which increase ^{15}N in plants (Schmidt *et al*., 2005*;* Bahar *et al*., 2008).

Determination of various minerals in meat has been shown to be efficient tools for geographic origin authentication of meat. Some interesting results were obtained on poultry, lamb and beef meats (Bahar *et al*., 2008). To be conclusive, it could be assumed that each area of production exhibits a specific profile in some minerals. However, these methods suffer of serious limitations. First, several areas in the world have similar geological undergrounds. Second, some feeds such as cereals and protein sources are commercialized on a worldwide market. Third, some minerals are added in diets of animals through mineral complementation. That is why a multi-elemental analysis coupled with multivariate statistical analysis is required to ensure a good discrimination of geographical origin (Franke *et al*., 2005).

III – Rearing conditions

Rearing conditions (outdoor/indoor, age at slaughter, length of fattening) and feeding systems largely affect pig adipose and muscle tissues. These effects are marked in traditional pig production based on local breed (Iberian, Corsican, Basque …) and fattening diet relied to local food (acorns, chestnuts, grass). Numerous papers describe the chemical traits of pig adipose and muscle tissues as related to many parameters of rearing and feeding in both industrial and traditional pig production (ref). Some of these parameters are of great interest to trace rearing and feeding systems because they are highly variable: lipid content, fatty acid and triacylglycerol composition. Other minor components found in animal tissues, mainly in adipose tissue, are typical of feed source: vitamins, poly-phenols, and hydrocarbons (Prache *et al*., 2007).

1. Breed or genotype

Recent developments in biotechnology open a new field in the traceability and authentication of individuals, lines, genotypes and breeds. The biotechnological methods have been developed very fast for the last 20 years. In theory, these methods are able to give a genetic fingerprint indentifying perfectly each individual and permitting to trace each animal from farm to fork because DNA is specific to each individual. However, the cost of these methods is up to now too high for a routine use (Dalvit *et al*., 2007; Lockley et Bardsley, 2000).

In contrast, these tools should be very helpful to check the genotypes used for dry cured hams production in PDO where specifications refers to local breed or allows some crossbred genotypes (i.e. Duroc X Iberian) and bans industrial pig genotypes. The development of genetic tools for local breed authentication and their crossbred require a large data base including the typical traits of the main breeds and genotypes used in European pig production. Tracing the local breeds is crucial to the survival of the herds and to defends and valorizes the high quality dry cured hams. Several studies have been devoted to differentiate the Iberian pig breed and line and to control the level of Duroc blood in the crossbreds for detecting mislabeled dry cured hams (Alvez *et al*., 2002; Fernandez *et al*., 2004; Ovilo *et al*., 2000; Garcia *et al*., 2006). The tools are based on DNA microsatellites and AFLP fragments allow a good differentiation of Iberian from crossbred Duroc X Iberian but are less efficient to distinguish crossbred Duroc x Iberian (50/50) from these with a lower proportion of Duroc blood.

2. Feeding systems

As mentioned above, some stable isotope ratios such as $^{15}N/^{14}N$ and $^{13}C/^{12}C$ are good tracers of feeding systems. Thus $^{13}C/^{12}C$ ratio in meat is related to the proportions of C4 and C3 plants in the diet. In Europe, the main C4 plant used in animal feeding is maize which is included in the diet to increase energy density in feed. So an increased $^{13}C/^{12}C$ in meat is an indicator of a more intensive feed system (Bahar *et al*., 2005; Boner et Förstel, 2004). Similarly, an increased $^{15}N/^{14}N$ ratio in meat is related to an intensive system of feed production because this increase is related to more intensive use of fertilizers. These isotope ratios give interesting results in discriminating ruminant meat fed grass versus maize or reared onto organic system versus more intensive system (Piasentier *et al*., 2003). In Iberian pig, $^{13}C/^{12}C$ ratio is higher in adipose tissue in pigs fed on traditional system (acorns and grass) than in pigs fed on more intensive system (concentrate). The higher is the proportion of concentrate, the higher is $^{13}C/^{12}C$ ratio in the tissue (Gonzalez-Martin *et al*., 1999). This measurement of this ratio could be interesting for discriminating Bayonne hams from these produced in the other areas because maize is largely included in the feed of Bayonne pigs.

3. Lipid composition of adipose and muscle tissues

Lipids and lipid fractions have been often used to distinguish animal according to their rearing conditions. Thus it was established that fatty acid composition of both adipose and muscular tissues is strongly related to these of feeds in pigs because it is a monogastric animal. This is of particular interest to distinguish pigs fed on local feeds such as acorns, chestnuts or grass from these fed on concentrate. Regarding fatty acid composition of raw material, genotype is also a major factor of variation. In Europe, the higher quality dry cured hams are produced from local breeds with a slow growth rate which deposit large amount of fat during the fattening period when they are too old to deposit muscle. Consequently lipids contain a high proportion of monounsaturated fatty acids coming from the conversion of starch from diet into saturated and monounsaturated fatty acids. Both fatty acid and triacylglycerol compositions were used to distinguish pigs according to their breed (Local breeds versus crossbred) or their diet (local feeds versus concentrate). In all the cases, triacylglycerols are more efficient to discriminate pigs because small variations in fatty acid composition are correlated to large variations in triacylglycerol composition (Riaublanc *et al*., 1999). Several authors have succeeded to distinguish Iberian pigs according to the feeding systems based on fatty acid composition of lipids from adipose tissue, intramuscular fat or liver (Flores *et al*., 1988; Ruiz *et al*., 1998; Perez-Palacios, 2009) or on triacylglycerol composition of adipose or muscular tissues (Diaz *et al*., 1996; Tejeda *et al*., 2002; Viera-Alcaide *et al*., 2007). Some minor lipid components can be good indicators of local feed consumption. In various amounts in feeds, they are stored in body fat. Hydrocarbon profiles of adipose tissue were used to distinguish Iberian pigs according to feeding systems. n- alkanes are not efficient (Tejeda *et al*., 2001a) but some peculiar hydrocarbon such as eut-kaurene (Navaez-Rivas *et al*., 2008) and neophytadiene (Tejeda *et al*., 2001b, Perez-Palacios, 2009) coming from grass could be used to discriminate Montanera pigs fed on acorns and grass from other Iberian pigs fed various amounts of concentrate. Tocopherols, namely gamma one, which is in a high amount in acorns (Tejerina *et al*., 2010), could help to discriminate traditional Montanera feeding system from others containing concentrate (Perez-Palacios, 2009*;* Tejerina *et al*., 2010).

4. Age at slaughter

The age of pigs is regarded as one of the main parameters improving meat quality and is included in the specifications of dry cured hams in many areas of production. Up to now, no method allows tracing this physiological parameter.

IV – Processing

The changes in raw matter during dry cured ham processing are largely involved in the typical sensory traits of end products. These changes involved a complex set of chemical and physic-chemical reactions affecting lipids, proteins, water and salt contents (Gandemer, 2002*;* Toldra et Navarro, 2002, Toldra, 2006).. The intensity of these changes largely depends on the conditions of processing used in the main area of production in Europe. Many PDO specifications contain specific requirements on the different steps of the process (length, temperature)(Flores, 1997). The changes in chemical and physic-chemical traits of meat and adipose tissues of hams have been largely described and marked differences were observed according to methods of processing. However, very few papers focus on methods to check that the specific requirements on process written in specifications are respected.

The use of thawed meat is prohibited in high quality dry cured ham production. Several papers are devoted to the differentiation of fresh and thawed raw meat. A review of methods indicates that only a combination of several methods allows discriminating fresh from thawed meat including DNA degradation, enzyme profile in juice extracted from meat and microscopy techniques (Ballin and Lametsch, 2008).

Volatile profiles of dry cured hams depend on the length and the temperature of the main steps of the process as well as the chemical traits of the raw material. Many papers described differences in volatile profiles from hams of different countries or feeding systems (Ruiz zt al., 1999*;* Bolzoni *et al*., 1996*;* Dirinck *et al*., 1997). Some volatiles found in aroma of hams come directly from feeds and are tracers of feeding systems. However, quantification of volatiles is very difficult and results vary greatly according to the method of volatile extraction and from one laboratory to another. So, volatile analyses are not proper tools to discriminate hams.

V – Conclusion

This review shows that the research on the authentication of meat and meat products is in progress. This is a major concern for consumers and producers. However, very little has been done on dry-cured hams. So up to now, we lack of accurate methods to assess that the specifications of dry-cured hams produced under PDO, PGI ou CSC in Europe are strictly applied. Most of methods remain potential tools and are far from their use as standard recognized methods to detect mislabeled products. That is why most of these methods were developed with small sets of samples of well-known origins. These methods must be validated using large numbers of samples of unknown origins and processes including raw meat and dry-cured hams arising from intensive systems of production all around the world. Large opened data bases must be built putting together all the characteristics of dry-cured hams as related to their area and specifications of production. A better characterization of the environment where animals are reared is required to able to mobilize very promising methods based on stable isotope ratios or trace elements quantifications.

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Chemical composition of dry ham "*Kraški pršut*" predicted by NIR spectroscopy

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Abstract. Dry ham "*Kraški pršut*" is a Slovenian traditional meat product with protected geographical designation. This protection implies that certain *consortium* constrains and control of dry-cured ham quality should be respected. For regular checks NIR spectroscopy offers an interesting alternative to conventional chemical methods. The aim of the present study was to test the ability of NIR spectroscopy to predict several chemical constituents of "*Kraški pršut*". Proximate analysis (moisture, salt, protein, non-protein nitrogen, intramuscular fat, free amino acids) was performed in muscles *biceps femoris* (n=135) and *semimembranosus* (n=135) of the final product. The quality of predictive models was assessed on the basis of the coefficients of determination (R^2_{CV}) of cross-validation and residual predictive deviation (RPD, *i.e.* the ratio between standard deviation of the reference data and standard error (SE_{CV}) of cross-validation). Highly reliable prediction results were obtained for moisture, protein, salt content and the percentage of salt per dry matter (R^2_{CV} > 0.90, RPD > 3.0). For intramuscular fat, free amino acids content and non-protein nitrogen reasonable calibration models were obtained (R^2_{CV} between 0.62 and 0.87, RPD between 1.6 and 2.8). Due to good prediction ability and the simplicity of measurement NIR spectroscopy offers good opportunity to replace time-consuming, expensive and/or hazardous laboratory methods.

Keywords. NIR spectroscopy – Ham – Chemical composition.

La composition chimique du jambon sec "Kraški pršut" prédite par la spectroscopie NIR

Résumé. *"Kraški pršut" est un jambon sec traditionnel slovène protégé par l'indication géographique. Cette protection implique la certification du produit et le respect de contraintes imposées par le consortium. Pour des contrôles réguliers la spectroscopie NIR offre une alternative intéressante pour remplacer les méthodes chimiques classiques. L'objectif de la présente étude était de tester la capacité de la spectroscopie NIR pour prédire plusieurs constituants chimiques du jambon sec "Kraški pršut". L'analyse chimique (humidité, sel, protéines, azote non protéique, gras intramusculaire, acides aminés libres) a été réalisée sur deux muscles du produit final, notamment le* Biceps femoris *(n = 135) et le semi-membraneux (n = 135). La qualité des modèles de calibrage a été évaluée sur la base de coefficients de détermination de la validation croisée (R^2_{CV}) et du coefficient RPD, à savoir le rapport entre l'écart-type des données de référence et l'erreur de la validation croisée (SE_{CV}). D'excellents résultats ont été obtenus pour la prédiction de l'humidité, des protéines, de la teneur en sel et du pourcentage de sel dans la matière sèche (R^2_{CV} > 0,90, RPD > 3,0). La précision de la prédiction du gras intramusculaire, de la teneur en acides aminés libres et de l'azote non protéique (R^2_{CV} entre 0,62 et 0,87, RPD entre 1,6 et 2,8) était un peu inférieure mais acceptable. En raison d'une bonne précision de la prédiction et de la simplicité des mesures, la spectroscopie NIR pourrait remplacer les méthodes chimiques plus laborieuses pour le contrôle régulier du produit.*

Mots-clés. *Spectroscopie NIR – Jambon sec – Composition chimique.*

I – Introduction

"*Kraški pršut*" is a traditional Slovenian dry-cured ham of the carstic region (called Kras in Slovenian language) which belongs to the family of Mediterranean type of dry-cured ham, characterised by dry salting, absence of smoking and long maturation. The product is economically important and highly appreciated among Slovenian consumers (Čandek-Potokar *et al*., 2004). On the national level "*Kraški pršut*" is protected by geographical designation and certified, implying that certain *consortium* constrains should be respected in regard to green ham properties, processing losses, chemical and sensory properties of dry-cured hams.

For regular checks near infrared (NIR) spectroscopy offers an interesting alternative over the conventional methods. Since the method enables fast and simple determination of many parameters simultaneously, it could effectively replace lengthy and expensive analyses which are less suitable to be used on a large scale and in particular for assays that are healthy and environmentally damaging (*e.g.* determination of intramuscular fat content based on hazardous organic solvent extraction). High potential of NIR spectroscopy for the prediction of chemical composition and the quality of raw meat was demonstrated in many previously published studies (for review see Prevolnik *et al*., 2004; Prieto *et al*., 2009). The literature reports relating to the application of NIR spectroscopy for the analyses of meat products are not abundant; the majority of them demonstrate good prediction ability (for review see Prieto *et al*., 2009). The lack of relevant studies is in particular evident in case of dry-cured ham. The few published studies on dry-cured ham were focused mainly on the prediction of sensory characteristics of dry-cured ham (Cruz Ortiz *et al*., 2006; García-Rey *et al*., 2005).

Due to the usefulness of NIR spectroscopy for mass analyses in the quality control of dry-cured ham the aim of the present study was to test the ability of the method to predict several chemical constituents of "*Kraški pršut*" dry-cured ham as this would be of high importance for the practice *i.e.* for regular checks in the production of certified product.

II – Materials and methods

1. Sampling and chemical analysis

The research was carried out on 135 dry-cured hams which were processed according to the rules of *consortium* for "*Kraški pršut*". The analysis is based on pooled data for two muscles, *biceps femoris* and *semimembranosus*. Prior to the chemical analyses, samples were trimmed of superficial fat tissue, cut in small pieces, frozen in liquid nitrogen, grinded to fine dust using a laboratory mill (IKA M120, IKA Werke, Staufen, Germany) and stored in plastic tubes at −20°C until further use. Chemical determinations (moisture, protein, intramuscular fat, non-protein nitrogen, salt and free amino acid content) were carried out in replicates.

For the determination of moisture content (ISO 6496, 1999), 5 g of the sample was mixed with equal amount of quartz sand and dried at 103°C to a constant mass. The loss of mass was recorded and expressed as a percentage of moisture in the sample.

For sodium chloride (salt) content determination (Monin *et al*., 1997), 1 g of sample was mixed with 80 ml distilled water and boiled at 100ºC for one hour. After cooling, 2 ml of 15% potassium ferrocyanide and 2 ml of 30% zync acetate was added and diluted with distilled water to 100 ml. After filtration, the NaCl content was determined by potentiometric titration using DL53 General Purpose Titrator (Mettler Toledo, Schwarzenbach, Switzerland). Additionally, salt content was expressed as the percentage of moisture or dry matter.

Protein content was calculated from total nitrogen content (ISO 5983-2, 2005) using the Kjeltec 2300 nitrogen analyser (Foss Analytical, Hileroed, Denmark). The organic matter in the samples was degraded by heating with concentrated sulphuric(VI) acid in the presence of catalysts. After the addition of base (NaOH) the resulting ammonia gas was dissolved in boric acid solution and titrated with hydrochloric acid. The total nitrogen content was calculated from the amount of the hydrochloric acid used for the titration. To obtain total protein content total nitrogen content was multiplied with 6.25.

For determination of non-protein nitrogen, 2.5 g of sample was homogenised in 25 ml of distilled water and centrifuged (Monin *et al*., 1997). Afterwards, 10 ml of 20% trichloroacetic acid was added, stirred well and let to stabilise for 60 min at room temperature. After the centrifugation, the supernatant was filtered and 15 ml of it used for determination of nitrogen in the same way as described for total nitrogen (ISO 5983-2, 2005). Additionally, non-protein nitrogen was expressed as a percentage of total nitrogen (proteolysis index).

Intramuscular fat content (ISO 1443, 2001) was determined using Büchi Extraction System B-811 (Büchi Labortechnik AG, Flawil, Switzerland). The samples were boiled with dilute hydrochloric acid to free the occluded and bound lipid fractions; the resulting mass was filtered and dried, the fat retained on the filter was extracted with light petroleum. The resulting fat was expressed as a percentage of fat in the sample.

The content of free amino acids was determined according to ISO 13903 (2005) adapted for dry-cured ham (internal laboratory protocol). Free amino acids were extracted with dilute hydrochloric acid. Co-extracted nitrogenous non-amino acid macromolecules were precipitated by adding sulfosalicylic acid and removed by filtration. The pH of the filtered solution was adjusted to 2.20. FAA were separated by ion exchange chromatography and determined by reaction with ninhydrin with photometric detection at 440 nm (for proline) and 570 nm (for other free amino acids) using Agilent 1200 series HPLC apparatus (Agilent technologies, Waldbronn, Germany) equipped with sodium cathion exchange column 8 μm, 3.0×250 mm (Pickering Laboratories, Mountain View, CA, USA), Pinnacle PCX post column derivatization instrument (Pickering Laboratories, Mountain View, CA, USA) and Agilent 1200 series Diode array and multiple wavelength detector (Agilent technologies, Waldbronn, Germany).

2. NIR spectra acquisition and spectral data analysis

Minced samples of analysed muscles were separately put in rectangular quartz cup (47×57 mm^2) about 3 mm thick, covered by paper disc and placed directly in NIRS apparatus. For each sample one scanning was performed. The samples were scanned with spectrophotometer NIR System model 6500 (Silver Spring, MD, USA) in a wavelength range from 400 to 2500 nm. Absorbance data were collected every 2 nm as log *1/R*, where *R* represents reflectance.

Spectral data processing was performed using WinISI II software. Calibration models were developed using modified partial least squares regression with internal cross-validation. Samples for which the difference between actual and predicted values exceeded 3 standard deviations (SD) were considered as outliers.

The mathematical treatment applied was 1 4 4 1, where the first number indicates the order of the derivative (1 is the first derivative of the *log 1/R*), the second number is the gap in nm over which the derivative is calculated, the third and fourth number refer to the first and the second smoothing. The “SNV and Detrend” option was used to correct scatter effects in the spectra. Within development of calibration models we tested spectral range (visible spectrum ranging from 400 to 1100 nm, NIR spectrum ranging from 1100 to 2500 nm and the whole spectrum ranging from 400 to 2500 nm). The number of PLS factors was limited to 16, but the actual number of PLS factors was defined separately for every single calibration model respecting the fall of cross-validation errors. The quality of calibration models was assessed through standard error of calibration (SE_C), coefficient of determination in calibration (R_C), standard error of cross-validation (SE_{CV}) and coefficient of determination in cross-validation (R_{CV}). Models’ performance was additionally evaluated using residual predictive deviation (RPD) which was calculated as the ratio between the SD of reference data and SE_{CV}.

III – Results and discussion

1. Material

Basic statistics of analysed chemical constituents in *biceps femoris* and *semimebranosus* muscles is presented in Table 1. Using two muscles we obtained broad range of variability which is of great importance for the development of calibration models.

2. Prediction of chemical composition

In the present study the same sample set was used to develop calibration models and to validate models. Namely, our previous studies (Čandek-Potokar *et al*., 2006; Prevolnik *et al*., 2009) showed that external validation (prediction) on the independent set of samples yielded comparable results as the cross-validation. In the present study prediction results are presented as statistical parameters of calibration (R_C and SE_C) and cross-validation (R_{CV} and SE_{CV}). The parameter RPD (the ratio SD/SE_{CV}) was also applied as an indicator of models' quality. Namely, RPD evaluates the prediction errors in view of the SD of the reference data and should be over three for accurate predictions (Andrés *et al*., 2008; Kennedy *et al*., 1996). Lower RPD values can be attributed either to a narrow variation range of the reference values (giving small SD) or to large NIR prediction error compared to SD of the reference values. Moreover, RPD enables to compare models' quality for the constituents/traits with different variation range where the prediction errors cannot be directly compared.

Table 1. Basic statistics for chemical constituents of dry ham (two muscles)

	Mean±SD	Range
Moisture, g/kg	537±51.4	435−652
Salt content, g/kg	71.2±8.19	44.8−94.1
Salt per DM, %	15.7±2.96	8.8−22.2
Salt per moisture, %	13.3±1.49	9.1−17.4
Protein, g/kg	34.2±5.08	26.0−43.9
NPN, g/kg	12.2±1.10	7.0−14.7
Proteolysis index, %	22.8±4.32	13.3−31.1
IMF, g/kg	38±14.0	16−88
FAA, mg/100g DM	7140±895	5237−9507

DM – dry matter; SD – standard deviation; IMF – intramuscular fat;
NPN – non-protein nitrogen; FAA – free amino acids.

As regards the prediction ability of NIR spectroscopy (Table 2), the results show, that there was a negligible difference in the prediction of chemical constituents based on NIR or the whole spectral range. Visible spectrum was also tested, but the results are not presented as the models showed considerably lower prediction accuracy. On the whole, R_{CV} for different chemical constituents ranged from 0.65 to 0.96 and RPD from 1.7 to 5.0. Highly reliable results were obtained for salt content and the percentage of salt per moisture or dry matter for which the R_{CV} were over 0.90 and RPD exceeded three which is indicative of highly reliable predictive models. Satisfactory results were obtained also for moisture, non-protein nitrogen and intramuscular fat content (R_{CV}=0.80−0.90, RPD=2.2−2.8), while for other constituents (protein, proteolysis index and free amino acids content) moderate results were obtained (R_{CV}=0.65−0.80, RPD=1.7−2.0).

Good calibration and cross-validation results in NIR (and consequently in the whole) spectral range can be explained by high correlations (up to ±0.80) between assessed chemical constituents and the absorbance data (data not shown) in wavelength range from 1100 to 2400 nm. In the dry-cured ham water represents 43-65% of the total fresh matter. Absorbance peaks of O-H bounds at 1450 and 1940 nm (Shenk *et al*. 1992) explain satisfactory NIR predictability for water content. Good performance of NIR spectroscopy to predict fat content is due to the strong absorption of C-H bonds in the NIR region at 1000 to 1400, 1700 and 2200-2400 nm (Shenk *et al*. 1992). Regarding protein, specific absorbance of N-H bonds could be found in the NIR region from 1460 to 1570 nm and from 2000 to 2180 nm (Shenk *et al*. 1992). In the case of proteins, it should be mentioned that protein content was calculated on the assumption that all nitrogen in the sample appears in protein, although a part (27%) of nitrogen is in form of non-protein nitrogen. High correlation coefficients between average spectrum and salt content led to good prediction results although NIR spectroscopy is known to be unable to detect inorganic substances unless they are bound to organic substance (Van Kempen, 2001). It is likely that in dry-cured ham salt content is indirectly predicted from other compounds (*e.g*. correlation coefficient between salt and water content amounts to 0.53). Moreover, NaCl itself shows no absorbance in the NIR region, but the presence of dissolved salts gives rise to the wavelength shifts in the spectrum. This phenomenon has been used to assess the content of NaCl in meat products (Downey and Hildrum, 2004).

Table 2. Prediction of chemical composition using NIR spectroscopy in two dry ham muscles (*biceps femoris* and *semimembranosus*)

Constituent	Spectrum nm	n	Mean±SD	emin	emax	SE_C	R_C	SE_{CV}	R_{CV}	RPD	PLS
Moisture, g/kg	400-2500	262	538.7±51.6	383.9	693.6	15.8	0.91	16.3	0.90	3.17	4
	1100-2500	258	539.7±51.3	385.9	693.6	15.6	0.91	16.1	0.90	3.18	4
Salt, g/kg	400-2500	254	71.69±7.68	48.66	94.72	2.45	0.90	2.55	0.89	3.01	5
	1100-2500	253	71.70±7.69	48.63	94.77	2.28	0.91	2.36	0.91	3.25	5
Salt per DM, %	400-2500	257	15.81±2.88	7.17	24.44	0.85	0.91	0.91	0.90	3.15	5
	1100-2500	254	15.85±2.89	7.17	24.53	0.86	0.91	0.91	0.90	3.16	5
Salt per moisture, %	400-2500	254	13.33±1.41	9.10	17.56	0.46	0.89	0.50	0.87	2.80	6
	1100-2500	255	13.33±1.40	9.12	17.55	0.50	0.87	0.52	0.86	2.69	5
Protein, g/kg	400-2500	264	33.96±5.05	18.81	49.12	1.42	0.92	1.57	0.90	3.22	7
	1100-2500	262	33.90±5.01	18.86	48.94	1.46	0.91	1.58	0.90	3.18	6
NPN, g/kg	400-2500	260	12.25±0.99	9.29	15.21	0.53	0.71	0.57	0.66	1.72	6
	1100-2500	258	12.25±0.97	9.34	15.17	0.57	0.65	0.59	0.64	1.65	5
Proteolysis index	400-2500	262	22.91±4.26	10.12	35.69	1.57	0.86	1.69	0.84	2.52	6
	1100-2500	262	22.92±4.25	10.17	35.66	1.55	0.87	1.70	0.84	2.50	8
IMF, g/kg	400-2500	259	36.6±12.3	0.0	73.7	7.5	0.63	7.6	0.62	1.63	3
	1100-2500	259	36.6±12.3	0.0	73.6	7.6	0.62	7.7	0.61	1.60	3
FAA, mg/100g DM	400-2500	258	7120±879	4484	9757	348	0.84	382	0.81	2.30	6
	1100-2500	258	7124±873	4507	9742	396	0.79	410	0.78	2.13	4

DM – dry matter; NPN – non protein nitrogen; IMF – intramuscular fat content; FAA – free amino acid content; SD – standard deviation of the reference values (calculated after the elimination of outliers); emin – estimated minimum; emax – estimated maximum; SE_C – standard error of calibration; R_C – coefficient of determination of calibration; SE_{CV} – coefficient of determination of cross validation; R_{CV} – standard error of cross validation; RPD – residual predictive deviation (ratio SD/se_{CV}); PLS – number of PLS factors.

3. General discussion

Our results demonstrated high potential of NIR spectroscopy to predict chemical constituents and amino acid content of dry-cured ham which is very important for the industry to fulfil the *consortium* requirements. Presently, producers use classical/wet chemistry which is lengthy, expensive, often hazardous and thus less interesting to be used on a large scale. Since NIR spectroscopy enables fast and simple determination of many parameters simultaneously, it could effectively replace regular checking of dry-cured ham chemical constituent prescribed by the *consortium*.

In the literature there are a few literature reports on meat products (Collell *et al*., 2010; Cruz Ortiz *et al*., 2006; Gaitán-Jurado *et al*., 2008; García-Rey *et al*., 2005; González-Martín *et al*., 2009; Ortiz-Somovilla *et al*., 2007), moreover, there is a lack of information regarding the prediction of chemical composition of dry-cured ham. Accuracy of NIR predictive models obtained cannot be directly compared for different meat products because of different matrix, different constituents' variation range, *etc*. Literature reports (Gaitán-Jurado *et al*., 2008; Ortiz-Somovilla *et al*., 2007) on meat products (mainly pork sausages) showed successful prediction of fat, moisture and protein with NIR spectroscopy (R_{CV}=0.88-0.99, RPD=2.9-10.4). Excellent prediction results were published also by Collell *et al*. (2010) for moisture content in fermented pork sausages (R_{CV}>0.99, RPD>20). Similar as in our study they also managed to predict salt (NaCl) content with high accuracy (R_{CV}=0.97, RPD=6.2). Ellekjær *et al*. (1992) reported errors in the range of 0.4 to 1.3 g/kg in the prediction of salt content in cooked sausages.

IV – Conclusions

In the present work NIR spectroscopy proved as highly reliable method for the prediction of studied chemical constituents of dry-cured ham "*Kraški pršut*". For eventual replacement of (conventional chemical) methods currently used in regular checking of certified products the calibration models should be extended with samples of the whole a slice of dry-cured ham containing several muscles and adjacent fat.

Acknowledgements

The authors acknowledge the financial support from the state budget by the Slovenian Research Agency (for the program Agrobiodiversity P4–0072 and PhD scholarship of Maja Prevolnik).

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Evaluation of a handheld near infrared (NIR) spectrometer for the discrimination of Iberian pigs according to their feeding regime

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Abstract. The aim of this study was to evaluate a handheld near infrared spectroscopy (NIRS) instrument to classify Iberian pig carcasses in different commercial categories for instantaneous quality control and authentication in the industry. Sixty intact subcutaneous adipose tissue samples taken from the tail insertion area in the coxal region of the body were analyzed, twenty per category ("Bellota", "Recebo" and "Cebo"). A discriminant analysis based on the algorithm PLS2 was performed and different spectra pretreatment were evaluated. A Principal Component Analysis (PCA) showed more clear differences between "Cebo" samples and the other classes, while "Bellota" and "Recebo" were spectrally more similar within them. The external validation of the classification models based on a second derivative showed only one "Recebo" sample misclassified as "Bellota". These results show the high potential of the handheld NIRS device evaluated for the individual authentication of Iberian pig carcasses to discriminate the different feeding regime followed by the animal during the growing-finishing period.

Keywords. Iberian pigs – NIR Spectroscopy – Classification – MEMS spectrometer – Adipose tissue.

Évaluation d'un spectromètre portatif dans le proche infrarouge (NIR) pour la discrimination des porcs Ibériques selon leur régime

Résumé. *L'objectif de cette étude était d'évaluer un instrument portatif de spectroscopie dans le proche infrarouge (NIRS) pour classer les carcasses de porcs Ibériques selon les différentes catégories commerciales pour le contrôle de la qualité instantanée et de l'authentification dans l'industrie. Soixante échantillons de tissu adipeux sous-cutané intact prélevés dans la zone d'insertion de la queue dans la région coxale du corps ont été analysés, vingt par catégorie ("Bellota", "Recebo" et "Cebo"). Une analyse discriminante basée sur l'algorithme de PLS2 a été réalisée et les différents spectres de prétraitement ont été évalués. Une analyse en composantes principales (PCA) a montré plus de différences claires entre les aliments des échantillons "Cebo" et les autres classes, tandis que "Bellota" (gland) et "Recebo", spectralement, étaient plus semblables en leur sein. La validation externe des modèles de classification basés sur une dérivée seconde montre un seul échantillon "Recebo" classé à tort comme "Bellota". Les résultats montrent le potentiel élevé de l'appareil de poche NIRS évalué pour l'authentification individuelle des carcasses de porcs Ibériques, afin de discriminer les différents régimes d'alimentation suivis par les animaux pendant la période d'engraissement.*

Mots-clés. *Porcs Ibériques – Spectroscopie NIR – Classement – Spectromètre MEMS – Tissu adipeux.*

I – Introduction

The feeding regime of the animals during the final period of growing plays an important role in Iberian pig products quality (Garrido and De Pedro, 2007), within other factors that also influence such as the genotype (Ramírez and Cava, 2007), age or rearing conditions (Bonneau and Lebret, 2010). Those factors have a significant impact on the fatty acid composition and mainly in the high unsaturated/saturated fatty acid ratio which at the end is responsible of the exceptional organoleptic and healthy properties of the Iberian meat products (Cava *et al.*, 2000; Ventana *et al.*, 2007). The Spanish legislation classifies the animals into four commercial categories depending on the feeding regime and production system (BOE, 2007): "Bellota" (i.e. animals in free range fed exclusively with grass and acorns), "Recebo" (i.e. animals fed with acorns and grass supplemented with compound feeds in a outdoor system), "Cebo" (i.e. animals fed with compound feed in an intensive system) and "Cebo de campo" (i.e. animals fed exclusively with compound feed in free range).

Official classification methods (on-farm inspections and gas chromatography of melted fat) are high cost and time-consuming, not providing individual items but batches information of animals groups that can have individual variability. Fast, accurate, objective, low cost and individual analysis methods are demanded by consumers and industry for quality control and authentication of these high market prices products. Since 1992, the potential of Near Infrared Spectroscopy (NIRS) has been highlighted for the analysis of melted fat (De Pedro *et al.*, 1992; García-Olmo *et al.*, 2009), intact adipose tissue (De Pedro *et al.*, 2007) and live Iberian pigs (Pérez-Marín *et al.*, 2009) as a tool for classifying Iberian pig animals into different commercial categories on the basis of the feeding regime. Few applications have been evaluated for on-line analysis (Perez-Marín *et al.*, 2009) and nowadays there are appearing in the market new handheld devices of low cost and easy analysis presentation, providing instantaneous results readily-available. In this study is evaluated a handheld micro electron mechanical system (MEMS) NIRS spectrometer for classifying Iberian pigs carcasses according to their feeding regime.

II – Materials and methods

1. Sample Set

Sixty Iberian pig adipose tissue samples were measured, taken from the tail insertion area in the coxal region of the body, where traditional gas chromatography biopsy is taken (De Pedro, 2001). The samples were stored at -20ºC until 24 hours before the NIRS analysis. The sample set was composed of 20 samples of each commercial category studied ("Bellota", "Recebo" and "Cebo"). Animal feeding regime was controlled by trained personnel.

2. NIRS measurements

A handheld MEMS-NIRS instrument (Phazir 2400, Polychromix Inc., Wilmingon, MA, USA) was used to collect reflectance spectra in the range 1600-2400 nm with a resolution of 8 nm (resolution-pixel 8nm, resolution-optical 12nm). A quartz protection was used for preventing dirt accumulation in the instrument. Three spectra per sample were collected and the mean spectrum per sample was used for further analysis.

3. Data modelling

As spectral pre-treatments, Standard Normal Variate (SNV) plus Detrending (DT) (Barnes *et al.*, 1989) was used to remove the multiplicative interferences of scatter and two derivative mathematical treatments were performed: window-wise filtering (1,10,5,1) and (2,5,5,1) (ISI, 2000). A Principal Component Analysis (PCA) was performed in order to detect spectral outlier samples and observe possible groups tendency. After outlier detection, the data set was divided in two: a training and validation (5 samples of each category) sets using the SELECT algorithm of the WinISI software. Discriminant analysis based on Partial Least-Squares (PLS2) was applied to classify the subcutaneous adipose tissue in the different commercial categories studied. The optimum number of model factors was selected by cross-validation using 4 groups. Those chemometric analyses were performed using the software WinISI II ver 1.50 (Infrasoft International, Port Matilda, PA, USA). The classification models were statistically evaluated, by calculating the number of animals correctly classified.

III – Results

Figure 1 shows the mean spectra of each commercial category analyzed. It was observed a similar pattern for all the groups, although it seems that there is a difference in absorbance range. Fat peaks were recorded at around 1720-1760, 2150 and 2310-2340 nm (Williams and Norris, 1987; Osborne, Fearn and Hindle, 1993); characteristic absorption bands at around 1940 nm were water-related (Williams and Norris, 1987; Osborne, Fearn and Hindle, 1993).

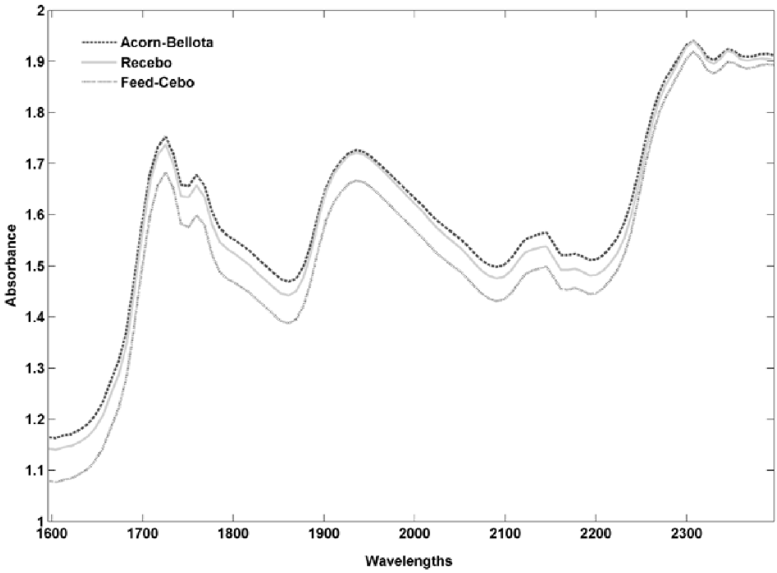

Fig. 1. Raw spectra (mean spectra) for each Iberian pig category studies.

PCA was performed to visualize the main structure of the data set and detect spectral outlier samples. It was observed 7 samples far from the centre of the population or with possible classification error due to the individual variability of the animals. After being removed, a new PCA analysis was performed using 9 Principal Components (PCs) explaining the 98.86%. The score plot (Fig. 2) shows clear differences between "Cebo" cluster and the other categories. "Recebo" and "Bellota" showed an overlap cluster, probably due to they are samples with a fatty acid profile more similar since those animals have eaten both acorns and grass.

Table 1 shows the statistics and number of samples correctly-classified of the training set, after outlier detection, for PLS discriminant. Two spectra pre-treatments were evaluated and a second derivative provided better classification results. A 91.66% of the "Bellota" samples were correctly classified, 63.6% of the "Recebo" samples and 100% of the "Cebo" samples. Table 2 shows the external validation of the model performed with a second derivative. Only one sample of the "Recebo" group was misclassified as an "Bellota" sample.

It should be remarked that any sample of the group "Cebo" (products with lower prices) was misclassified in the other groups ("Bellota" or "Recebo", products with higher prices) or vice versa. These results confirm the possibility to discriminate Iberian pig adipose tissue from animals reared under different feeding regime using a handheld MEMS-NIRS instrument that can be implemented in on-line applications in the industry.

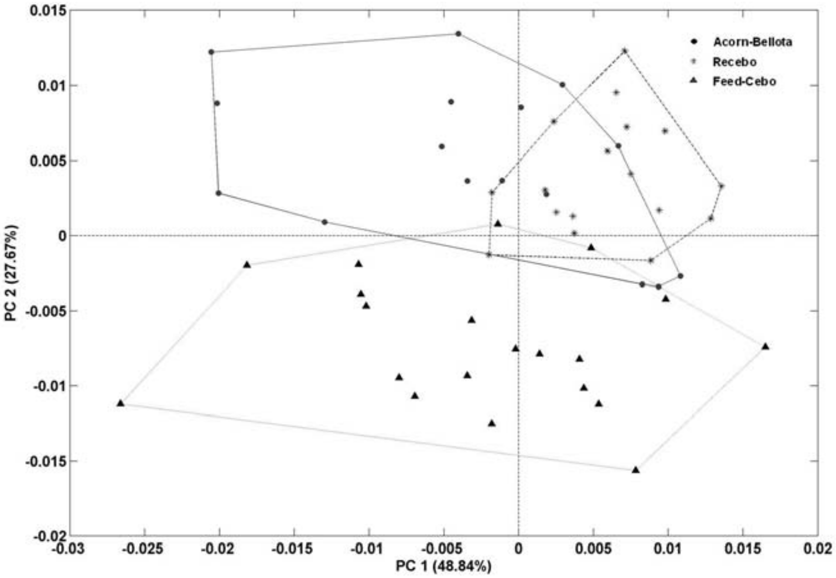

Fig. 2. PCA score plots for the two principal components.

Table 1. Classification results obtained by the PLS discriminant with a second derivative

Origin	No. of samples	Classified as		
		Bellota	Recebo	Cebo
Bellota	12	11	1	0
Recebo	11	4	7	0
Cebo	15	0	0	15

Table 2. External validation of the second derivative PLS discriminant model

Origin	No. of samples	Classified as		
		Bellota	Recebo	Cebo
Bellota	5	5	0	0
Recebo	5	1	4	0
Cebo	5	0	0	5

IV – Conclusions

The classification of Iberian pig intact adipose tissues using the handheld NIRS device evaluated was successful. PLS discriminant performed better classification predictions with a second derivative with only one sample of the "Recebo" group misclassified for the external validation. These results indicate the feasibility of performing an individual authentication of Iberian pig carcasses according to the feeding regime of the animals in a fast and with possibility of on-line applications. Further work is required in order to develop robust classification models with larger data sets and transfer the technology from laboratory to an on-line monitoring system.

Acknowledgements

The authors gratefully acknowledge the financial support provided by the INIA (Project number: RTA08-36) and the research projects of the Andalusian Regional Government AGR-285 and AGR-5129. EZR acknowledges financial support from the Spanish Ministry of Education as a fellow of the Program “Training of University Teachers” (Formación del Profesorado Universitario, FPU).

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Differential expression of sarcoplasmic protein in 'Casertana', 'Calabrese' and PEN AR LAN Pork

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Abstract. The characterization of the water soluble fraction of muscle proteins was carried out on 30 samples of meat taken from a pool of muscles (*Semimembranosus, Semitendinosus* and *Biceps femoris*) each representative of the 30 subjects {[20 ancient autochthonous genetic type (AAGT) with 'black' coat [10 '*Casertana*' (CT), 10 '*Apulo Calabrese*' (Calabrese) (CL)] and 10 PEN AR LAN ('white' coat, from cross breeding Large White and Landrace)}. Protein profile was analyzed by two-dimensional gel electrophoresis and MALDI-TOF mass spectrometry. The comparison of 60 two-dimensional maps was performed by Image Master 2D-Platinum software in order to establish the position and relative intensity, expressed as vol %, of each spot for each gel. In the range of our observation, image analysis showed 32 spots common to all samples analyzed; 17 spots of 32 common differed in relative 'abundance' (P<0.05). These spots, identified by peptide mass fingerprint, were classified as metabolic, cellular defense and other protein types. The results suggest a further possible use of proteomic approach in the tracing back of traditional food.

Keywords. MALDI-Tof fingerprint – Pig – Sarcoplasmic proteins – Two-dimensional gel electrophoresis.

Expression différentielle des protéines de la fraction soluble dans l'eau pour la viande des TGAA 'Casertana', 'Calabrese' et des hybrides commerciaux PEN AR LAN

Résumé*. La caractérisation de la fraction soluble des protéines musculaires a été effectuée sur 30 échantillons de viande provenant d'un pool de muscles (*Semimembranosus, Semitendinosus *et* Biceps femoris*) pour chacun des 30 sujets traités {20 de type génétique autochtone ancien (TGAA) à robe 'noire' [10 'Casertana' (CT), 10 'Apulo Calabrese' (Calabrese) (CL)] et 10 PEN AR LAN (à robe 'blanche', d'ascendants Large White et Landrace)}. Le profil protéique a été évalué en utilisant des procédures analytiques telles que l'électrophorèse bidimensionnelle couplée à la spectrométrie de masse MALDI-TOF. La comparaison des 60 cartes a été réalisée avec le software Image Master 2D-Platinum afin de comparer la position et l'intensité relative, exprimée en % vol de chaque spot pour chaque gel. Dans le cadre de l'observation, l'analyse d'image a mis en évidence 32 spots communs à la totalité des échantillons analysés ; 17 spots sur les 32 diffèrent en % vol (P<0,05). Ces spots ont été ensuite identifiés par peptide mass fingerprint et classés comme protéines du métabolisme, de défense cellulaire et autres protéines. Les résultats suggèrent une possible utilisation ultérieure de l'approche protéomique pour des études de caractérisation visant, entre autres, à l'analyse de la traçabilité.*

Mots-clés*. MALDI-Tof fingerprint – Porc – Protéines de la fraction soluble dans l'eau – Électrophorèse bidimensionnelle.*

I – Introduction

Proteomic analysis defines the identity, the structure and the relative abundance of proteins in a given cell type in a specific set of conditions. Proteins are the expression of genetic inheritance which also undergo post-translational modifications (phosphorylation, glycosylation and acetylation). The study of these modifications, by proteomic approach, may assist to differentiate animal species. The proteomic approach can be used for 'molecular characterization' of raw materials and their products; this 'characterization' can be employed in order to: (i) trace back product; (ii) point out flow chart phases; (iii) characterize possible relationships among protein (and their fragments) 'quantity' and 'quality attributes' of raw materials and their products.

The aim of this contribution was to suggest a proteomic approach to differentiate swine races. Differential analysis of the proteome in relation to the soluble fraction (sarcoplasmic) was carried out on meat samples taken from a pool of muscles (*Semimembranosus, Semitendinosus* and *Biceps femoris*) in pigs.

II – Materials and methods

The study involved 30 meat samples taken from a pool of muscles (*Semimembranosus, Semitendinosus* and *Biceps femoris*) each representative of the 30 subjects {[20 ancient autochthonous genetic type (AAGT) with 'black' coat [10 '*Casertana*' (CT), 10 '*Apulo Calabrese*' (Calabrese) (CL)] and 10 PEN AR LAN ('white' coat, from cross breeding Large White and Landrace)}. The slaughter was carried out in a single establishment and the carcass maturation was carried out in refrigerator at a temperature of 2-4 °C for a period of approximately 72 hours.

The analysis covered a total of 60 samples (2 for each sample) processed in parallel by:

(i) 2D-IPG-SDS-PAGE:

- the first dimension (IEF-IPG) was carried out by Ettan IPGphor II (GE Healthcare) using Immobiline DryStrips gel pH 3-10NL (18 cm) rehydrated with a solution of 8 M Urea, 0.5% CHAPS, 0.2% DTT, 0.5% IPG Buffer.
- the second dimension was carried out in accordance with the procedure of O'Farrell (1975) in polyacrylamide gradient gel electrophoresis (T = 9-18% and C = 2.5%) by using Ettan Twelve System (GE Healthcare).

(ii) Image analysis of two-dimensional maps (2-DGEm):

- was performed by software Image Master 2D-Platinum (GE Healthcare) quantifying in vol % the expression level of each spot; the spots found in common to three swine races were subjected to statistical analysis Student's t test.

(iii) Identification by MS:

- each spot was digested *in situ* with trypsin according to the procedure of Shevchenko *et al.* (1996) and tryptic digests were analyzed with Ettan MALDI-Tof/PRO mass spectrometer (GE Healthcare).

III – Results and discussions

The measurements carried out by MALDI-Tof mass spectrometer allowed us to identify 49 spots (Figure 1, Table 1). Some proteins appeared heterogeneous with differences in mass [MW (kDa)] and / or isoelectric point (pI) and they were identified as: Adenylate kinase, Myoglobin, Peroxiredoxin, Phosphoglycerate mutase, Similar parvalbumin, Transferrin, Triosephosphate isomerase. This heterogeneity could be due to:

(i) Genetic polymorphism (for example single nucleotide polymorphism).

(ii) Post-transcriptional modifications (for example alternative splicing).

(iii) Post-translational modifications (for example glycosylation and phosphorylation).

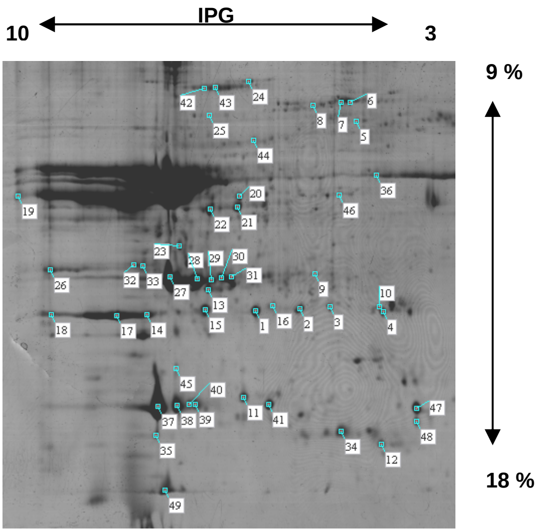

Fig. 1. 'Casertana' AAGT. Two-dimensional map of the sarcoplasmic proteins.

Considering the three breeds, 32 spots (65% of those identified) resulted in common to totality of subjects analyzed. The proteins identified were grouped by protein function as reported in Table 1:

(i) Metabolic protein:

- adenylate kinase (it catalyzes the reversible transfer of terminal phosphate group between ATP and AMP),
- enolase (it is an enzyme involved in the development and regeneration of striated muscle).

We compared 32 common spots of the three swine races analyzed. The differences observed were analyzed by Student's t test (Figure 2; Table 2) and divided in significant (P<0.05-0.001) and near to significant (P<0.20-0.10):

- CL *vs* CT: 14 comparisons out of 32 [44%, 6 of which significant (P<0.05-0.001) and 8 near to significance (P<0.20-0.10)].
- CL *vs* PEN AR LAN: 22 comparison out of 32 [69%, 11 of which significant (P<0.05-0.001) and 11 near to significance (P<0.20-0.10)].
- CT *vs* PEN AR LAN: 16 comparison out of 32 [50%, 12 of which significant (P<0.05-0.001) and 4 near to significance (P<0.20-0.10)].

Table 1. Spots identified by MALDI-Tof mass spectrometry. Red spots were common to all two-dimensional maps

Group	Spot, N	Protein name
Cellular defense proteins	1, 2, 3	DJ-1
	4	Glyoxalase
	5	HSP 60
	6, 7, 8	HSP 70
	9, 10	Peroxiredoxin
	11	Superoxide dismutase
	12	Thioredoxin
Metabolic proteins	13, 14, 15, 16, 17, 18	Adenylate kinase
	19	Aldolase
	20, 21, 22	Enolase
	23, 24	Muscle Creatine kinase
	25, 26	Phosphoglicerate mutase
	27, 28, 29, 30, 31	Triosephosphate isomerase
Transport proteins	32, 33	Carbonic anhydrase
	34	H-FABP
	35	Haemoglobin ß-chain
	36	Haptoglobin
	37, 38, 39, 40, 41	Myoglobin
	42, 43	Transferrin
Indicators of proteolysis	44	Leucine aminopeptidase
	45	Muscle Creatine kinase fragment
Miscellaneous	46	Zinc finger protein
	47, 48	Similar Parvalbumin
	49	Similar to polyubiquitin

The major number of significant and near to significant critical limit comparisons was found in the comparison CL *vs* PEN AR LAN (69%) while the lower number (44%) was found in the CL *vs* CT comparison. It is interesting to note that the major differences in protein expression were detected among race with 'white' coat (PEN AR LAN) and those with 'black' coat (CL and CT). The differences related to the proteins classified by function as:

(i) cellular defense proteins (DJ-1, HSP 70, thioredoxin),

(ii) metabolic proteins (adenylate kinase, enolase, triosephosphate isomerase),

(iii) transport proteins (H-FABP, haemoglobin beta chain, myoglobin),

(iv) proteolysis indicator [leucine aminopeptidase (LAP)],

(v) other proteins (parvalbumin).

Table 2. Differentially expressed spots. Comparison between 'Apulo Calabrese' (Calabrese), 'Casertana' and PEN AR LAN. Identification by MALDI-Tof mass spectrometry

Group	Spot, *N*	Protein name	CL *vs* CT	CL *vs* H	CT *vs* H
Cellular defense proteins	2	DJ-1	<<<	***	***
	3		**	***	***
	4	Glioxalase	<	<<<	
	5	HSP 60	<<<		<<<
	6	HSP 70		<	
	7			<<	**
	8			<<	*
	12	Thioredoxin	**	<<	
Metabolic proteins	13	Adenylate kinase	**	<<<	
	14		<<<		*
	15		<<<	<<<	
	20	Enolase		<<<	<
	21			*	<<<
	22			*	<<<
	23	Muscle creatine kinase	<<<	<<<	
	27	Triosephosphate isomerase	*	*	
Transport proteins	34	H-FABP	<<	***	**
	35	Haemoglobin ß-chain		**	*
	37	Myoglobin		***	**
	38		*	***	**
	39		***	***	**
Indicators of proteolysis	44	Leucine aminopeptidase	<<	<<	*
Miscellaneous	47	Similar Parvalbumin		*	<<

CL = 'Apulo Calabrese' (Calabrese); CT = 'casertana'; H = Pen Ar Lan 'Hybrid'

< P<0.20; << P<0.15; <<< P<0.10; * P<0.05; ** P<0.01; *** P<0.001

Our results highlighted the importance of 'race' for relative quantitative expression (volume, %) of spots considered, as previously showed in sheep and pig by Matassino *et al*. (2010 a,b), and for energy metabolism that may be different depending from coat color ('black' or 'white'). In fact, in the 'white' pig the higher expression of enzymes involved in the glucidic metabolism and the lower expression of myoglobin may point out that muscle cells favour the glycogen catabolism and, so, in order to balance the energy metabolism, they may pre-eminently use glucose rather than fatty acid. The opposite behaviour is observed in 'black' pig in which, an increased expression of myoglobin and H-FABP and a decreased expression of glucose metabolism enzymes would indicate a metabolism mainly based on oxidative chain. This is also confirmed by the predominantly ‘oxidative’ nature of some muscles of 'black' and 'belted' pigs caused by a higher percentage of Slow Oxidative fibers (SO), as evidenced by previous research (Matassino *et al.*, 1993; Barone *et al.*, 2000, 2005). This would result in a different use of the energy stored by two genetic types of pigs: the 'black' pig would use preferentially energy stored in adipose tissue while the 'white' pig would use energy stored under glycogen form. This difference in the metabolic activity of muscles may be attributed to a different use of the metabolic - energy pathways (glycolysis or oxidative). This differentiation could derive from a diversity of both genetic (for example coat color gene) and hormonal by stress (for example adrenaline) nature; both could influence the structure and the functionality of the muscular fiber.

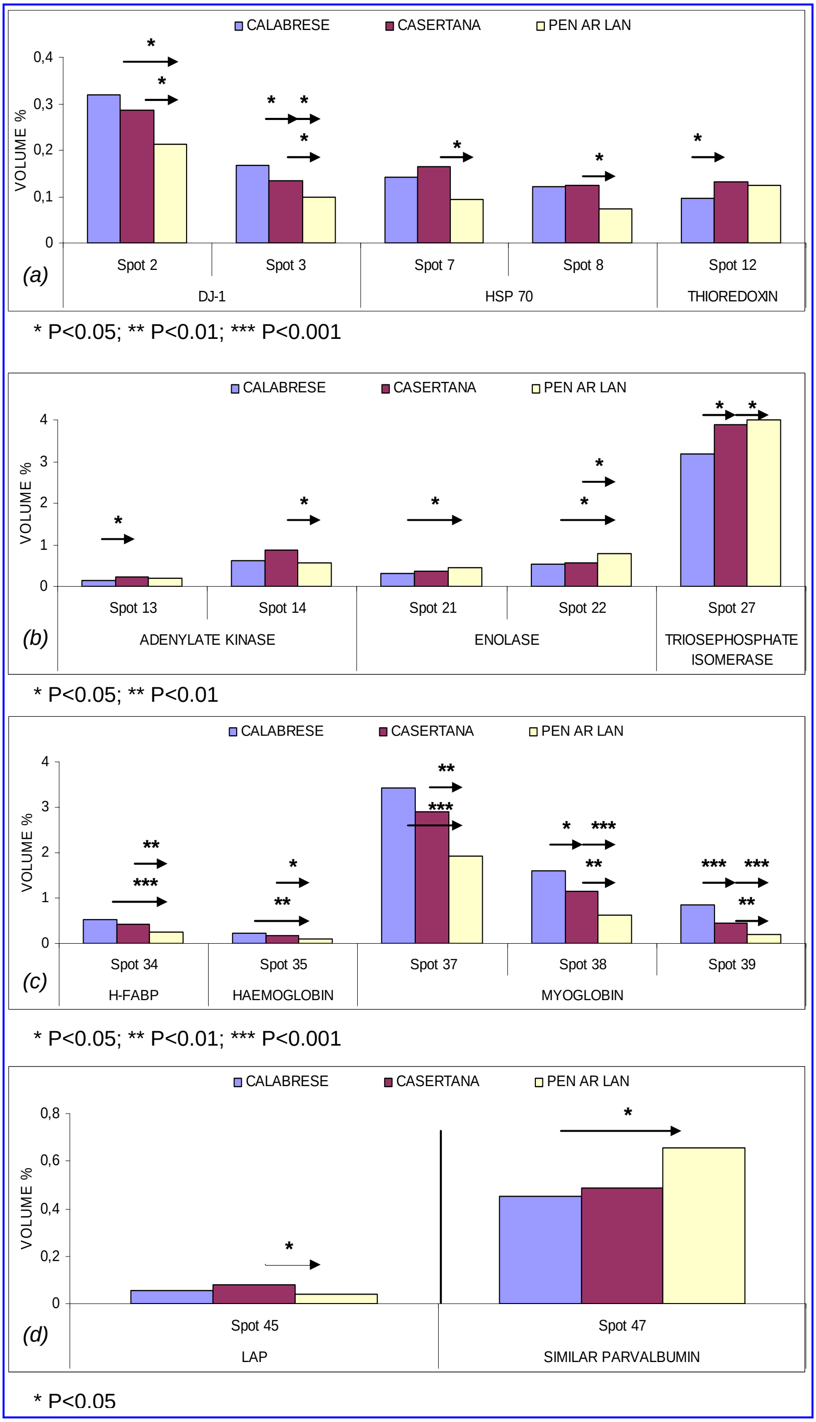

Fig. 2. Spots differentially expressed grouped in: (a) cellular defense proteins, (b) metabolic proteins, (c) transport proteins, (d) indicator of proteolysis and miscellaneous.

This differentiation could derive from a diversity of both genetic (for example coat color gene) and hormonal by stress (for example adrenaline) nature; both could influence the structure and the functionality of the muscular fiber.

These results demonstrated that it was possible to study muscle phisiology not only by fiber composition but by molecular level too.

The increased expression of cellular defense proteins in 'black' pig may suggest a better its response to phenomena of environmental stress. This is in agreement with the greater ability to constructivism (Matassino,1989; Lewontin, 1993) hypothesized for ‘black’ pig in comparison with ‘white’ pig, being the former also in a initial phase of anthropic conditioning.

IV – Conclusions

The proteomic approach to the study of sarcoplasmic protein of muscle revealed the differences in energy metabolism between 'black' and 'white' pig. These differences could represent sources of 'molecular characterization' that could allow both to trace back meat of different genetic types and to assess their 'nutritional' and 'extra-nutritional' quality. However, it is necessary to extend sampling and to deepen the study of muscular protein composition.

Acknowledgements

The research was supported by Ministry of Agricultural, Food and Forestry Policy (MiPAAF) and by Selmol project.

The Authors want to thank dressed pork factory ’Di Maria’ – Cese Alte – Circello (BN).

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Is it possible the breed origin traceability of Iberian pigs?

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Abstract. Two different approaches for the verification of *Iberian* breed origin, in both animals alive and meat products, are jointly described in this study. The former is based on two genes that present polymorphisms with alleles that are exclusive (*MC1R*4*) or present high frequencies (*IGF2* g.3072A) in *Duroc* breed and are absent in *Iberian*. The use of these markers allows to discriminate Iberian purebred from crossbred *Duroc* x *Iberian* but it does not fully detect pigs with a lower proportion of *Duroc* genes. The second approach is centered on the use of single nucleotide polymorphisms (SNP) presenting divergent frequencies in both breeds. After hybridization with PorcineSNP60 BeadChip of samples from different *Iberian* (26) and *Duroc* (15) origins, we selected 96 SNPs with differences of allelic frequencies larger than 0.8 and evenly distributed over the 18 autosomes. Simulations were carried out estimating that 48 out of these SNPs would allow the verification of breed origin, with errors ranging from 1% to 4%, both in purebred and in crossbred animals with different *Duroc* proportion. Two diagnostic analyses based respectively on 750 and 230 samples genotyped for the panel of 96 SNPs have been performed with different purposes and satisfactory results.

Keywords. Breed traceability – Iberian – Duroc – SNP.

La traçabilité de l'origine des porcs de race Ibérique est-elle possible ?

Résumé. *Deux approches sont décrites pour vérifier l'origine de la race Ibérique, applicables aux animaux vivants ainsi qu'à leurs produits dérivés, grâce à l'utilisation de marqueurs génétiques. La première est basée sur l'utilisation de deux gènes qui présentent des polymorphismes avec des allèles fixés (MC1R*4) ou qui présentent une fréquence élevée (IGF2 g.3072A) chez la race Duroc et qui sont absents chez la race Ibérique. L'utilisation de ces marqueurs permet de discriminer les génotypes Ibériques purs par rapport aux animaux issus de croisements Duroc x Ibérique, mais ne détecte pas complètement les porcs ayant un fond génétique Duroc moins important. La deuxième approche est basée sur l'utilisation des polymorphismes de type SNP, présentant des fréquences alléliques différentes chez les deux races. Après hybridation des échantillons de différentes origines Ibériques (26) et Duroc (15) sur la puce de génotypage PorcineSNP60 BeadChip, nous avons sélectionné 96 SNPs parmi ceux qui présentaient des différences inter-races de fréquences alléliques supérieures à 0,8 et qui étaient répartis sur les 18 autosomes. Les simulations réalisées estiment que l'utilisation de 48 de ces SNPs permettrait la répartition par race des animaux purs et croisés avec Duroc dans des proportions différentes, en obtenant des taux d'erreur compris entre 1% et 4%. Nous avons réalisé deux analyses diagnostiques basées sur le génotypage de 750 et 230 échantillons, respectivement, sur le panel de 96 SNPs, avec des objectifs différents et des résultats satisfaisants.*

Mots-clés. *Traçabilité – Ibérique – Duroc – SNP.*

I – Introduction

Iberian pigs are the source of highly priced meat and dry-cured products. The optimum quality of meat and hams is associated with both the combination of purebred Iberian genotypes and traditional extensive fattening called *Montanera*. However Iberian pigs are commonly crossbred with Duroc animals, and even other dark coated breeds like *Large Black*, to improve their efficiency for lean growth. The Spanish regulation of 'Iberian' labelling only admits progenies from Iberian sows and boars crosses with males from other breeds than *Duroc*. Moreover cured products are labelled as '*Ibérico*' or '*Ibérico Puro*' depending if they proceed from *Duroc* x *Iberian* crossbred or from purebred *Iberian* animals. In the case of products to be exported, additional quality controls could be eventually carried out by customer countries. Full traceability is increasingly being demanded by producers and consumers but traditional tagging systems presents several difficulties, mainly in *Iberian* pigs, because of their extensive management system (López-Bote, 1998).

In this context, genetic markers could be a useful tool to check the breed origin of living animals and products as well as to verify the parentage relations registered on the Herd Book. Microsatellite markers have been proposed for estimate the genetic composition of dry-cured *Iberian* hams (García *et al*., 2006). However this approach requires the genotyping of a high number of markers (>25) and preliminary analyses on parental populations are needed to determine their allelic frequencies pattern. In addition, these markers can be difficult to score and are not amenable to automation. Single nucleotide polymorphisms (SNPs) present several advantages over microsatellite markers: higher abundance in the genome, more easily to handle and interpret in laboratory and better compatibility with automation. Although SNP are usually bi-allelic and consequently less informative than microsatellites, this disadvantage can be overcome by genotyping a higher number of SNPs. Our group has been working in the search of *Iberian* or *Duroc* exclusive genetic markers and, despite the close genetic relationship between these breeds, some exclusive alleles were reported on nuclear (Fernández *et al*., 2004) and mitochondrial (Alves *et al*., 2009) genes. Indeed analysis of *MC1R* and *IGF2* alleles are already in use and mitochondrial DNA markers could result particularly useful for *Iberian* maternal origin validation. The problems arise when samples to analyze carry less than 50% of *Duroc* genes. In these cases neither of the available tests does itself results enough to certify genetic origin (Rodriguez-Ramilo *et al*., 2008). Advances in high-throughput DNA sequencing and genotyping have led to the recent commercialization of a high density porcine SNP array. The aim of this work was to use the 60K porcine SNP array to develop a low-density panel of evenly spaced SNPs and check its effectiveness for differentiate purebred *Iberian* from crossbred pigs with a wide range of *Duroc* genes.

II – Materials and methods

1. High-density genotyping

In a first step, 41 samples were analyzed including 15 *Duroc* boars from 12 different genetic origins and 26 *Iberian* pigs (both males and females) from 16 breeding nuclei registered in the Herd Book. Genomic DNA was extracted from blood samples according to the standard phenol-chloroform method or from ear tag biopsies using the PureLink™ Genomic DNA kit (Invitrogen, Spain). All these samples were genotyped for 62,163 SNPs, using the Porcine SNP 60 BeadChip (Illumina, San Diego, CA, USA). Genotyping reactions were performed on an "Infinium DNA Analysis Assay" at the Veterinary Service of Molecular Genetics (Universitàt Autonoma de Barcelona, Spain).

2. SNP selection and evenly spaced low-density genotyping

The porcine SNP60 BeadChip features 62,163 evenly spaced probes with an estimated one marker per 40Kb across the pig genome. Genotyping data were analyzed and those SNPs that failed to produce an amplification product, that have no information about their location or map over sexual chromosomes were eliminated of the study, yielding a total of 45,180 SNPs. A first SNP selection was based on (*i*) their informativity i.e. the SNP that presented divergent frequencies between the two analyzed breeds with between-breed differences of allelic frequencies larger than 0.80 and (*ii*) regular distribution over the 18 autosomes. We also incorporated additional probes in order to check the feasibility of genotyping highly informative SNP for breed origin verification (*IGF2*, *MC1R*, *OCA2*) not included in the Porcine SNP 60

BeadChip. The Illumina Assay Design Tool (ADT) was used for evaluating individual loci and creating the most successful custom genotyping assay. The 96 SNP loci finally selected were simultaneously interrogated with the GoldenGate Genotyping Assay. Two analyses were performed with different goals and based respectively on: *a*) 750 DNA samples of purebred pigs of both breeds and probationary *Iberian* pigs, and *b*) 230 DNA samples from *Iberian* sows and *Duroc* boars and controlled F1 *Duroc* x *Iberian* crossbred pigs.

3. Simulations and statistical analysis

Previous simulations were performed assuming different number of bi-allelic SNPs (24, 48 and 96) with diverse values (from 0.60 to 0.80) for the allelic differences between *Iberian* and *Duroc* breeds. In each simulation, one hundred reproducers of each breed were simulated with the correspondent number of SNP genotypes. From the alleles of reproducers of each breed we sampled two hundreds F1 crossbred pigs, and a similar procedure was used between purebred and F1 individuals to simulate four hundreds pigs of each one of the backcrosses: F1 x *Iberian* and F1 x *Duroc*. According to its profile of allelic frequencies, each one of the simulated pigs was assigned to one of five clusters using an algorithm similar to STRUCTURE (Pritchard *et al*., 2000). One hundred replicates were obtained for each simulated case.

For the analysis of actual genotyping data software *Bayesian Analysis of Population Structure* (BAPS) v5 was used (Corander *et al*. 2003).

III – Results and discussion

1. SNP selection from High-Density Genotyping

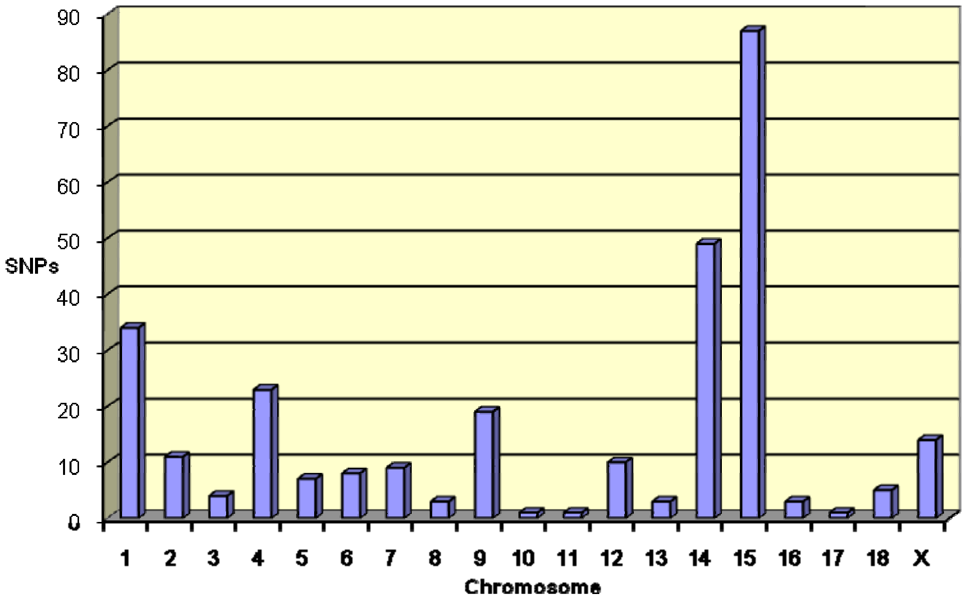

Fig. 1. Distribution over the porcine chromosomes of SNPs showing differences between breeds of allelic frequencies larger than 0.80, obtained from high-density genotyping with SNP60 BeadChip.

The genotyping of 15 *Duroc* and 26 *Iberian* samples with the porcine SNP60 BeadChip revealed a total number of 292 SNPs with allelic frequencies differences larger than 0.80 and mapped on different chromosomes (Fig. 1). The number of SNPs displaying divergent allelic frequencies is proportional to the total number of probes interrogated in each chromosome but it is not related with the size of the chromosome. That is the reason why the highest number of SNPs was observed on SSC14 and SSC15 that are smaller than SSC13. Despite this inconvenient, we tried to carry out the SNP selection taking into account the chromosomes size whenever possible and choice SNPs evenly spaced across the porcine genome. Moreover, as it was said before, we checked the feasibility of genotyping highly informative SNPs for breed origin verification (*IGF2*, *MC1R*, and *OCA2*) not included in the Porcine SNP 60 BeadChip. Assay Design Tool (Illumina) provided satisfactory scores for *OCA2*, *MC1R*2* and *MC1R*4* but not for *IGF2* g.3072A polymorphism. Inclusion of mitochondrial polymorphisms also had to be discarded because GoldenGate Genotyping Assay does not allow the genotyping of this kind of variation. The final set of 96 SNPs included 92 probes selected from the Porcine SNP 60 BeadChip, distributed over the 18 autosomes as follows: 11 on SSC1, eight on SSC2, three on SSC3, six on SSC4, six on SSC5, seven on SSC6, five on SSC7, three on SSC8, seven on SSC9, one on each one of SSC10 and SSC11, six on SSC12, four on SSC13, eight on SSC14 and also on SSC15, two on SSC16, one on SSC17 and five on SSC18. Moreover four additional probes were included, two for *MC1R*2* and one for each one *MC1R*4* and *OCA2*.

2. Simulation results

Results of two of the performed simulations were summarized in Tables 1 and 2. The first case corresponds to the use of a panel of 48 SNPs with remarkable allelic differences between breeds ($|q_{\text{IBERIAN}} - q_{\text{DUROC}}| > 0.80$), and the second to a larger panel of 96 SNPs with lower divergence of frequencies ($|q_{\text{IBERIAN}} - q_{\text{DUROC}}| > 0.60$). In both situations, the method performs very well for assigning adequately each individual to its correspondent genetic group. The observed error rates in these simulations ranged from 1% to 4% using 46 very divergent SNPs (Table 1), and from 0.1 % to 2% using 96 moderately divergent SNPs (Table 2).

Table 1. Allocation average proportions (over 100 replicates) to the different groups of simulated purebred and crossbred pigs: *Iberian* (*n* = 100), *Duroc* (*n* = 100), F1 *Duroc* x *Iberian* (*n* = 200), F1 x *Duroc* (*n* = 400) and F1 x *Iberian* (*n* = 400) genotyped for 48 SNPs with allelic differences between breeds greater than 0.80

	Iberian	Duroc	F1 Duroc x Iberian	F1 x Duroc	F1 x Iberian
Iberian	0.992	0.000	0.000	0.000	0.008
Duroc	0.000	0.992	0.000	0.008	0.000
F1 Duroc x Iberian	0.000	0.000	0.960	0.011	0.029
F1 x *Duroc*	0.000	0.002	0.033	0.965	0.000
F1 x Iberian	0.005	0.000	0.034	0.000	0.961

However, some of the assumptions of the simulations favour these positive results: *a)* purebred and crossbred pigs are related, the last ones being progenies of the *Iberian* and *Duroc* reproducers considered in the analyses, *b*) the number of clusters (five) is known and the possible hidden substructure of the purebred populations is ignored, and *c*) all the genotypes are available without missing marker data. The assumptions *a*) and *c*) are clearly unrealistic in practice, and the assumption *b)* is questionable at least for the Iberian breed where a hidden substructure has been previously inferred (Alves *et al*., 2006). The considered proportions of *Duroc* genes could be directly applied to the situation of Iberian products that usually proceed from these crosses. However, a lower proportion of the *Duroc* genome could be present at animals qualified as purebred *Iberian*. These arguments indicate that the ability of these techniques to solve the practical problems of traceability of *Iberian*-type live pigs and meat products will be lower than the expected according to the simulated results, and the use of panels of at least 96 very divergent SNPs ($|q_{IBERIAN} - q_{DUROC}| > 0.80$) seems advisable.

Table 2. Allocation average proportions (over 100 replicates) to the different groups of simulated purebred and crossbred pigs: Iberian (n = 100), Duroc (n = 100), F1 Duroc x Iberian (n = 200), F1 x Duroc (n = 400) and F1 x Iberian (n = 400) genotyped for 96 SNPs with allelic differences between breeds greater than 0.60

	Iberian	Duroc	F1 Duroc x Iberian	F1 x Duroc	F1 x Iberian
Iberian	0.999	0.000	0.000	0.000	0.001
Duroc	0.000	1.000	0.000	0.000	0.000
F1 Duroc x Iberian	0.000	0.000	0.972	0.009	0.019
F1 x Duroc	0.000	0.004	0.020	0.976	0.000
F1 x Iberian	0.005	0.000	0.015	0.000	0.980

3. Low-density genotyping

The finally selected panel of 96 SNPs was used for genotyping two sets with 750 and 230 DNA samples, respectively, pursuing different objectives. A verification of the purebred *Iberian* origin was performed on a set of analyzed samples proceeding from 82 *Iberian* pigs, 61 *Duroc* and 607 uncertain purebred *Iberian* pigs named here as probationary animals. Fig. 2 represents a graphic summary of the results obtained from one of the performed analysis: admixture based on pre-defined clustering. Purebred pigs from the two breeds were considered as the origin populations and the proportion of alleles proceeding from these populations was estimated for the probationary samples. The presence of *Duroc* genes was observed in different proportions for some of the probationary animals and also for six of the assumed purebred *Iberian*. However, more satisfactory results were obtained from admixture analysis based on the mixture clustering of individuals (results not shown). This analysis assumes the maximum uncertainty about the samples and besides of a cluster grouping the *Duroc* pigs, the clustering of individuals reveals a likely hidden substructure of three different clusters for all the samples (668) of possible *Iberian* origin. The posterior admixture analysis indicates that only a 2.5% of these samples present *Duroc* genes with a probability lower than 0.05.

As it was already mentioned the set of 96 SNPs included *MC1R*4* and *MC1R*2* probes that can supply additional information for problematic breeding nucleus. Besides, an additional genotyping of *IGF2* g.3072A/G was carried out (according to the procedure described by Van Laere *et al*., 2003), for samples proceeding from the herds where *Duroc* genes were detected in putative Iberian animals. Both *MC1R*4* and *IGF2* g.3072A are exclusive alleles of the *Duroc* breed. Their presence in animals from the same breeding nucleus evidence introgression of *Duroc* genes. Moreover, the presence of *MC1R*2* allele was detected in a few animals which indicates introgression of *Large Black* in those herds.

The goal of the second trial was to validate the *Duroc* x *Iberian* origin of commercial crossbred pigs. The 230 analyzed samples corresponded to seven *Duroc* boars, 50 *Iberian* sows and 173 controlled F1 *Duroc* x *Iberian* animals. In this case, admixture analysis based on pre–defined clustering was performed. The *Duroc* boars and the *Iberian* sows were considered as the source of genes of the crossbred animals, and the proportion of genes proceeding from each one of them was estimated for each individual. The obtained results are summarized on Fig. 3. We could infer the presence of about a 10% of *Duroc* genes on four out of the 50 assumed purebred *Iberian* sows, with a probability lower than 0.001. Results also confirm the crossbred origin of all the *Duroc* x *Iberian* pigs. However, for some of the crossbred animals the inferred percentage of *Duroc* genes was slightly different than the 50% expected. It can be explained by the low size of the origin breed samples that cannot represent all the within population diversity of these breeds for the selected genetic markers.

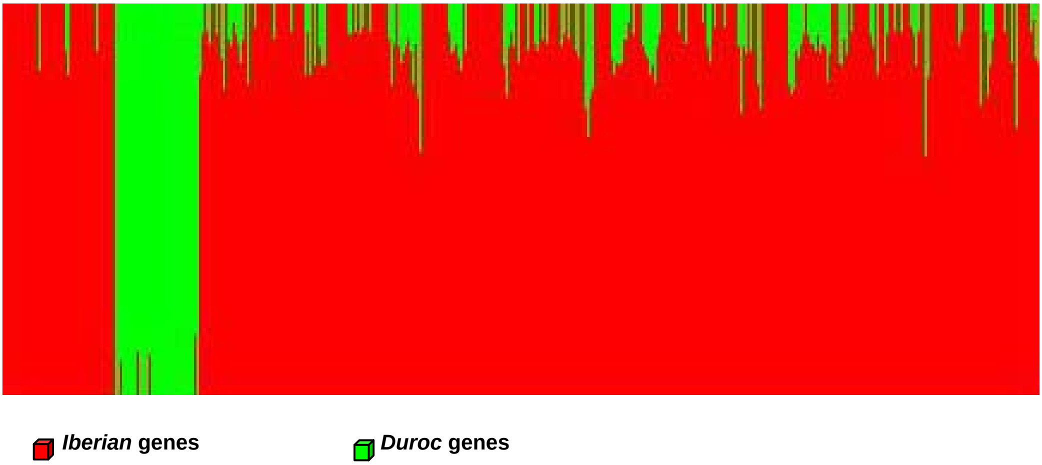

Fig. 2. Proportion of Iberian and Duroc genes on the 750 analyzed samples inferred from admixture based on pre-defined clustering.

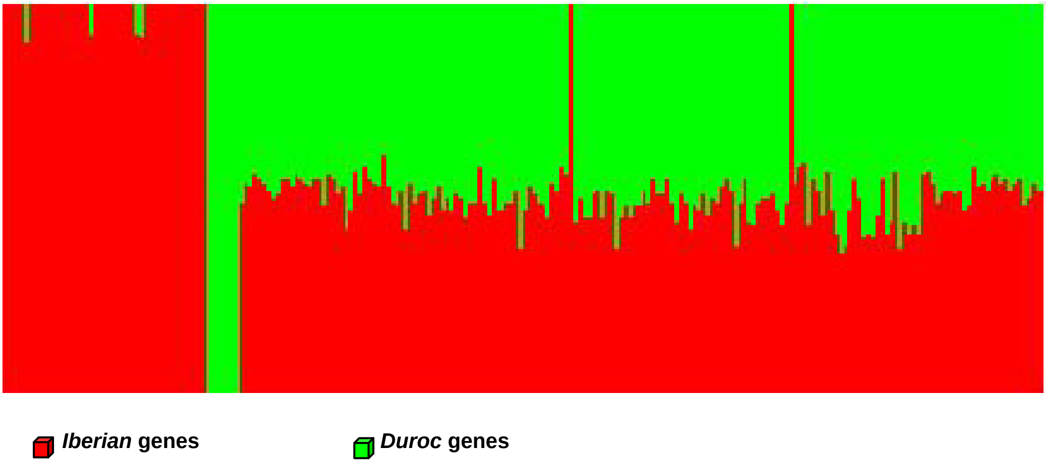

Fig. 3. Proportion of Iberian and Duroc genes on the 230 analyzed samples.

It was mentioned before that the selection of SNPs was based on the ADT SNPScore file output that allows include those assays that are predicted to have a high likelihood of success. However this does not guarantee the complete amplification of all the SNPs because low-density genotyping uses VeraCode technology whereas high-density genotyping uses Infinium technology. Hence, we observed that a number of probes, ranging from 10 to 13 failed to produce amplification. Moreover, taken into account the inclusion of two *MC1R**2 probes on the final set of 96 SNPs the actual number of useful probes varied between 82 and 85 instead the 96 selected SNPs.

IV – Conclusions

This study exemplifies how the recent advances in SNP discovery and high throughput automated genotyping methods can be applied to solve problems of authentication of genetic origin of *Iberian* pigs and their products. Low density genotyping of a moderate number of SNPs (< 90), with divergent frequencies between the *Iberian* and *Duroc* breeds, may be a powerful tool either to infer the purebred *Iberian* origin, to detect animals with low proportion of *Duroc* genes or to validate the *Duroc* x *Iberian* origin of commercial pigs or meat products. However, further research need to be carried out in order to build adequate databases for improving the usefulness of this procedure. We have planned to study a higher number of purebred and crossbred pigs, and to extend the panel of markers with new SNPs discovered using the next generation sequencing technology (Ramos *et al*., 2010).

Acknowledgements

The authors wish to acknowledge Juan García-Casco (AECERIBER) and Mario Gómez (Agroibéricos DeRaza) for providing samples, and technical assistance of Anna Mercadé (UAB). Financial support was received from the INIA CON09-006 and MICINN PET2008-0090 grants.

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Transcriptome analysis of the Iberian pig

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Abstract. Iberian pig meat is a remarkable constituent of the healthy Mediterranean diet. The organoleptic and nutritional properties of Iberian pig products are influenced by both genetic and environmental factors including the animal's diet. To investigate the potential impact of genotype and diet on gene expression eventually controlling taste and texture of the meat, we performed an in-depth transcriptomics analysis of the Iberian versus Duroc pig muscle from differentially fed animals. Muscle tissue samples were frozen in liquid nitrogen immediately after sacrificing the animals and stored at –80 ºC. Total RNA was extracted, retrotranscribed to cDNA and subjected to SuperTag Digital Gene Expression (ST-DGE) functional genomics analysis. The generated sequences were then counted and annotated to entries in public databases to assign potential functions to the expressed genes. To this end we identified significant breed-specific as well as diet-specific expression profiles of known as well as from hitherto unknown genes, involved in metabolic pathways related to the quality of Iberian meat products. These results demonstrate the power of genomics in general and transcriptomics in particular to identify differentially expressed profiles and putative candidate genes for quality control, certification and traceability, helping breeders and farmers to produce both animals and derived products (like ham) with higher nutritional value and improved organoleptic properties, to further enhance the healthy Mediterranean diet.

Keywords. Genome – Functional genomics – mRNA – Bioinformatics.

L'analyse du transcriptome du porc Ibérique

Résumé. *La viande de porc Ibérique est un constituant remarquable de la saine alimentation méditerranéenne. Les propriétés organoleptiques et nutritionnelles des produits du porc Ibérique sont influencées par des facteurs génétiques et environnementaux, y compris l'alimentation de l'animal. Pour étudier l'impact potentiel du génotype et du régime alimentaire sur l'expression génique du goût et éventuellement le contrôle et la texture de la viande, nous avons effectué une analyse en profondeur du transcriptome du muscle du porc Ibérique par rapport aux porcs Duroc à partir d'animaux nourris différemment. Des échantillons de tissus musculaires ont été congelés dans l'azote liquide immédiatement après l'abattage des animaux et conservés à -80º C. L'ARN total a été extrait, rétrotranscrit en ADNc et soumis à une analyse génomique fonctionnelle pour l'expression des gènes moyens selon la technique SuperTag (ST-DGE). Les séquences ont alors été comptées et annotées pour leur entrée dans des bases de données publiques pour assigner des fonctions potentielles aux gènes exprimés. À cette fin, nous avons identifié des profils d'expression significatifs spécifiques de la race ainsi que de l'alimentation, pour des gènes connus ainsi qu'inconnus jusque-là, impliqués dans les voies métaboliques liées à la qualité des produits de la viande du porc Ibérique. Ces résultats démontrent la puissance de la génomique et de la transcriptomique en général, et en particulier pour l'identification des profils différentiellement exprimés et des gènes candidats putatifs pour le contrôle, la certification de la qualité et la traçabilité, afin d'aider les éleveurs et les agriculteurs à produire des animaux et des produits dérivés (comme le jambon) ayant une valeur nutritionnelle plus élevée et de meilleures propriétés organoleptiques, pour améliorer encore le sain régime méditerranéen.*

Mots-clés. *Génome - Génomique fonctionnelle – ARNm – Bioinformatique.*

I – Introduction

The quality is an important parameter of any food product. This is particularly relevant for products with a Protected Designation of Origin (PDO) certification issued by the European Union (EU). Such label corresponds to food products that are specific of a particular region, conveying a particular quality or characteristic which is peculiar of such designated area. The PDO certification requires the food traceability from the origin to the consumer. Likewise, the National Quality Standard (NQS; "Norma de Calidad") for the Iberian pig products (ham, shoulder blade and loin) in Spain regulates the labeling of such food ("Real Decreto" 1469/2007, of 2nd November). Different approaches have been developed and deployed for food certification and traceability, being particularly efficient and convenient the ones based on DNA molecular markers. Yet, the development of such markers requires the prior knowledge of the DNA sequences differentiating the products to be identified and tracked down.

On the other hand, consumers are demanding with increasing emphasis not only quality-certified food products, but also –and specially– healthy products. The Iberian pig products are remarkable constituents of the healthy Mediterranean diet, which has been included in the United Nations Educational, Scientific and Cultural Organization (UNESCO) list of "Intangible Cultural Heritage of Humanity" since 18th November 2010. The healthy implications of the Iberian pig products have not been completely elucidated, but some studies have shown that their balanced compositions of unsaturated lipids protect against cardiovascular diseases (CVD), effectively reducing the plasma levels of Low Density Lipoprotein (LDL) cholesterol, total cholesterol, triglycerides and fibrinogen (Rebollo *et al.*, 1998; Martín *et al.*, 2009; Jiménez-Colmenero *et al.*, 2010).

Remarkably, the fat composition of the Acorn-Fed Iberian Ham (AFIH) has a surprisingly high content of oleic acid (monounsaturated lipid) for a product of animal origin (50 to 60% of the total fat), depending on the tissue considered. Thus, on the coccyx fat it reaches up to 57% (about 60% monounsaturated lipids), whereas in the intramuscular fat the oleic acid represents 45 to 50% and the saturated fats about 40%. The high oleic acid content of the Iberian ham resembles the one of the olive oil, which ranges from 55.0 to 83.0% of oleic acid content depending on cultivar. In this respect, it should be emphasized that the olive oil has been granted the label of "Qualified Health Claim" by the Food and Drug Administration (FDA) of the United States of America, due to its protective effect against CVD.

The organoleptic, nutritional and healthy properties of the Iberian pig products are influenced by both genetic and environmental factors including the animal's diet (eg., the acorn feeding, as previously indicated; the acorn has a similar lipid composition to the olive oil itself). To investigate the potential impact of genotype and diet on gene expression eventually controlling taste, texture and composition of the meat, we have performed an in-depth transcriptomics analysis of the Iberian *versus* Duroc pig muscle from ham of differentially fed animals. The final goals of this research are both to unravel the genomic, transcriptomic and metabolomic pathways that set apart the Iberian pig from other breeds, as well as to identify differentially expressed genes that can be used as tools for the certification and traceability of Iberian pig products.

Explanatory note: The purpose of this work is not to carry out a statistical analysis of many samples because: i) this kind of genomic experiments use a different methodological approach in the literature; and ii) the prohibitive cost of such statistical approach on genome-wide experiments, not being cost-effective. On the contrary, the purpose of this work is to carry out a genome-wide transcriptomic analysis in order to discover candidate genes that are highly differentially expressed (upregulated or downregulated) between the breeds and feedstuffs used. Such candidate genes are now being subjected to other analysis (validation using many animals from different herds), but such work is out of the scope of the present report. The readers interested on the methodology used in genomic studies are directed to the abundant bibliography on such matter for further details, as previously indicated.

II – Materials and methods

Female Iberian pigs from the most abundant lineage (Retinto) were selected from the same brood and fatten up for 70 days with either strict "montanera" (acorn-based with grass), standard cereal-based feedstuff without acorn (as well as female Duroc, used as control) or standard cereal-based feedstuff enriched with olein (oleic-acid-rich feedstuff). The standard feedstuff contained (mg/kg): barley (14.75×10^4), wheat (50.00×10^4), corn (21.00×10^4), soybean meal (1.20×10^4), lard (1.10×10^4), calcium carbonate (8.00×10^3), calcium phosphate (1.15×10^4), sodium chloride (4.00×10^3) and corrector (5.25×10^3). The olein-supplemented feedstuff contained (mg/kg): barley (47.50×10^4), wheat (15.00×10^4), bran (15.50×10^4), soybean meal (8.50×10^4), pulped beet (5.75×10^4), high-oleic sunflower meal (5.75×10^4), calcium carbonate (9.50×10^3), calcium phosphate (7.00×10^3), sodium chloride (4.00×10^3), corrector (5.00×10^3) and vitamin E mix (250 ppm). The Table 1 summarizes the three feedstuffs used.

Table 1. Feedstuff composition

Composition (%)*	"Montanera"		Standard	Olein
	Acorn	Grass		
DM	54.70	10.60	85.62	89.91
DM ash	2.11	13.54	3.89	4.56
DM crude protein	7.97	21.85	16.78	16.94
DM crude fiber	1.82	20.00	3.28	5.67
DM fat	7.06	3.07	3.55	8.54
DM NFE	80.50	46.60	72.57	64.35
SFA	12.12	21.75	25.15	11.15
MUFA	64.65	7.34	32.08	67.50
PUFA	21.60	70.92	42.68	21.52

(*): DM: Dry Matter; NFE: Nitrogen-Free Extracts; SFA: Saturated Fatty Acids; MUFA: Monounsaturated Fatty Acids; PUFA: Polyunsaturated Fatty Acids.

The pigs were transported to the slaughterhouse the day before slaughtering, trying to minimize the stress of the animals. Carbon dioxide was used for stunning just before bleeding, according to the specifications outlined in the European Union legislation. The Iberian pigs were slaughtered after 14 months old (150 to 170 kg); the Duroc pig grows faster and thus was slaughtered after 9 months old (157 kg).

The ham muscle tissue samples (*biceps femoris*; femoral biceps) were frozen in liquid nitrogen immediately after sacrificing the animals, and stored at –80 ºC until needed. The total RNA was isolated from samples using Trizol from Life Technologies (Carlsbad, CA, USA). In short, the tissue was ground under liquid nitrogen with a pestle and mortar. A small volume of ground tissue was dissolved in 1 ml Trizol and homogenized. The RNA was isolated after the addition of 1/5 volume of chloroform. Subsequently, the RNA was precipitated with isopropanol, washed and dissolved in diethylpyrocarbonate-treated water. The RNA concentration was determined using a NanoDrop 2000 (Thermo Scientific, Waltham, MA, USA). The RNA quality (integrity) was checked by agarose gel electrophoresis. The RNA was retrotranscribed to cDNA with reverse transcriptase and subjected to SuperTag Digital Gene Expression (ST-DGE) functional genomics analysis (Anisimov, 2008; Datson, 2008; Matsumura *et al.*, 2008a,b, 2010; Wang, 2008; Zaretzki *et al.*, 2010). The generated sequences were then counted and annotated to entries in public databases to assign potential functions to the expressed genes.

III – Results and discussion

As previously indicated, the main purpose of this work is not to carry out a statistical study with many animals, but to use the currently validated genomic methodological approaches to carry out a genome-wide transcriptomics analysis. The usefulness of such strategy to identify differentially expressed genes and thus putative candidate genes for further applications is widely supported in the literature. More than a million Expressed Sequence Tags (EST) have been isolated and sequenced, including differentially expressed ones between breeds and feeds. Thus, we have identified breed-specific as well as diet-specific expression profiles of known as well as from hitherto unknown genes, some of them being involved in metabolic pathways related to the characteristics of Iberian meat products.

The transcriptomic profiling of Iberian pigs fed with either acorn, standard feedstuff or olein were compared with the transcriptomic profiling of Duroc pigs fed with standard feedstuff. A total of 15,814 to 18,952 expressed tags were compared for each pair of breeds and feeding, generating gene expression fold changes from +14.58 to +18.09 on one side and –13.71 to –16.58 on the other side. This represents a remarkable wide range of variation.

The analysis of such data has allowed the identification of differentially expressed genes (eg., involved in lipid and peptide metabolism and transport), besides genes that are currently unknown. The expression of genes related with lipid biosynthesis was higher on Iberian *vs* Duroc pigs. Likewise, for acorn-fed versus standard-fed Iberian pigs. These results indicate that both the genotype as well as the diet determine the higher lipogenic gene expression in the acorn-fed Iberian pig.

These results demonstrate the power of genomics in general, and transcriptomics in particular, to identify differentially expressed genes between pig breeds and feeding conditions. The data generated will contribute to a better understanding of the pig transcriptomics and metabolomics pathways. These results are also being exploited and validated analyzing many individuals from different herds (data not shown, corresponding to future publications) to design DNA molecular markers for quality control, certification and traceability. These developments will help breeders and farmers to produce both animals and derived products (like ham) with higher nutritional value and improved organoleptic properties, to further enhance the healthy Mediterranean diet. Likewise, they can contribute to increase the consumer confidence for products with higher quality and price, which can further assist the promotion and consolidation of demanding markets (Japan, USA, etc).

Acknowledgments

Supported by “Consejería de Agricultura y Pesca” of “Junta de Andalucía” (013/C/2005), “Grupos PAI” AGR-158 and AGR-248, and “Universidad de Córdoba” (“Ayuda a Grupos”), Spain. We thank the “Cooperativa del Valle de los Pedroches” (COVAP; Pozoblanco, Córdoba, Spain) and the “Matadero Industrial de Cortegana S. A.” (MICSA; Cortegana, Huelva, Spain) for their assistance in obtaining samples.

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Classification of Iberian pigs as to their nutrition through the ChemSensor technique

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Abstract. We have developed a classification method to discriminate the quality of Iberian pigs with different feeding patterns (*bellota*, *recebo*, *extensive* and *intensive* feeding) in view of the large price differences encountered in the market depending on availability of acorns diet or other type of food. We used the *ChemSensor* Technique which combines a gas chromatograph with a mass detector as well as a liquid autosampler and a headspace system. Our starting material consisted of 207 samples of back side bacon extracted from pigs previously fattened with different feeding systems and subsequently fatty acid and volatile compound analysis were performed. Up to 75% of the samples were analyzed in a first step obtaining a prediction model that applies to the remaining 25% of samples. A proper software adds mass abundances in the range of m/z scanned (41-550) for further analysis by a chemometric software (Pirouette, by Infometrix Inc.). The classification method used was established with SIMCA (Soft Independent Modelling Class Analogy). Our results indicate a satisfactory classification up to 96% and a prediction of ca. 90% in bellota-type pigs and in intensive type and somewhat lower in recebo and extensive types.

Keywords. ChemSensor – Rating Iberian pigs – Mass Spectrometry – Traceability.

Classification des porcs Ibériques selon leur nutrition grâce à la technique du ChemSensor

Résumé. *Nous avons développé une méthode de classification pour distinguer la qualité du porc Ibérique élevé selon différents modes d'alimentation (*bellota*,* recebo*,* extensif *et* intensif*) en raison de grandes différences de prix sur le marché en fonction du régime alimentaire ou d'un autre type d'alimentation. Nous avons utilisé la technique ChemSensor qui combine un chromatographe en phase gazeuse avec un détecteur de masse ainsi qu'un échantillonneur automatique liquide et un système d'espace-de-tête. Notre matériau de départ se composait de 207 échantillons de lard arrière extraits de porcs engraissés avec différents systèmes d'alimentation et ensuite les analyses des acides gras et des composés volatils ont été effectuées. Dans un premier temps jusqu'à 75% des échantillons ont été analysés, obtenant ainsi un modèle de prévision appliqué aux 25% restants des échantillons. Un software ajoute l'abondance de masse dans la gamme des m / z numérisés (41-550) pour une analyse plus approfondie par un software de chimiométrie (Pirouette, par Infometrix Inc.). La méthode de classification utilisée était SIMCA (Soft Independent Modelling Class Analogy). Nos résultats indiquent une classification satisfaisante jusqu'à 96% et une prévision de presque 90% chez les porcs de type Bellota et de type intensif et un peu plus faible pour les types Recebo et les modes extensifs.*

Mots-clés. *ChemSensor – Evaluation du porc Ibérique – Spectrométrie de masse – Traçabilité.*

I – Introduction

The extensive production systems are appreciated because of the valuable preservation of natural environments as well as the well spread reputation of the meat products obtained. Spain is characterized by having a perfect ecosystem for such production, the *dehesa*, with over a million and a half hectares of cork and oak trees that supply the necessary food for the Iberian pigs. Acorns constitute a unique raw material for the Iberian pork industry, providing high valued products, like ham, paleta and pork loin. However, there is a high differential cost between this grazing system and the intensive, massive systems used by the bulk of farmers, which affects not only the cost of the animals themselves but the price of manufactured products.

The Spanish government has established quality standards classifying pigs as *bellota*, *recebo*, *extensive* and *intensive* types, depending on the final fattening phase.

There have been developed several analytical methods to assess this type of classification, one of them is presented here as part of a research proyect supported by the Instituto Nacional de Investigaciones Agrarias y Agroalimentarias (INIA) using the Agilent ChemSensor technique.

II – Materials and methods

Analysis were performed using a 4440B ChemSensor (*Agilent Technologies*), which combines a gas chromatography-mass spectrometer (GC / MS) with a chemometric software developed by *Infometrix Inc*. The former consists of a 6890 GC with a DB.23 column (*Agilent*), a liquid autosampler for 100 samples (HP 7683) and a 5973N MS mass spectrometer with a 5973 quadrupole detector operating in electron impact mode and scanning of masses in the m / z 41 and 550 range.

The starting material was backfat taken from Iberian pigs proceeding from some farms and livestock farms in Extremadura, Andalusia and Salamanca. The animals were fattened according to the *bellota*, *recebo*, *extensive cebo* and *intensive cebo* diet patterns. The total number of tested samples was 207, mostly pure Iberian breed, with 160 kg average weight. All the samples were originally obtained from the subcutaneous rump fat according to the standards (*ASICI specification*). After fat extraction using microwave and methyl transesterification with KOH in methanol, fatty acid methyl esters were recovered and submitted to the ChemSensor 4440B analyzer obtaining an array of ion mass fragmentation in the range m / z 41 and 550.

The whole number of samples (207 in total) was split as follows. In a first step the procedure involved an initial determination of the classification and predictive mathematical model based on known samples (156 samples, 75% of total) and in a second step it took place the prediction test, using a random set of samples (51 samples, accounts for *ca*. 25%).

III – Results and discussion

The chemometric analysis was applied using chemometric software SIMCA (*Soft Independent Modelling Analogy Class*) of *Infometrix Inc*. The first set of samples (156) was classified according to this model into four groups (see Table 1) with distances between groups shown in Table 2. We applied the above mathematical modelisation on the 25% remaining samples, obtaining the prediction results shown in Table 3.

Table 1. Classification results

	Classified				
Samples	***Bellota***	***Recebo***	***Cebo ext.***	***Cebo int.***	**% success**
Bellota	53	0	0	0	100
Recebo	0	53	1	0	98.15
Cebo ext.	2	1	24	0	88.89
Cebo int.	0	0	0	22	100
Total	55	54	25	22	97.44

Table 2. SIMCA model relative distances

	Distance to			
	Bellota	***Recebo***	***Cebo ext.***	***Cebo int.***
Bellota	0	0.9673	0.6596	3.1937
Recebo	0.9673	0	0.8810	3.6142
Cebo ext.	0.6596	0.8810	0	3.3592
Cebo int.	3.1937	3.6142	3.3592	0

Table 3. SIMCA model predictions

Actual samples	**Prediction**				
	Bellota	***Recebo***	***Cebo extensive***	***Cebo intensive***	**% success**
Bellota	16	0	2	0	88.8
Recebo	1	13	1	1	81.2
Cebo ext.	2	5	2	0	22.2
Cebo int.	1	0	0	7	87.5

IV – Conclusions

The use of ChemSensor for classification purposes proves to be highly reliable in all types of samples, grouped in four classes (*bellota, recebo, cebo extensivo, cebo intensivo*) with an average selectivity greater than 97%.

With the classification model generated with *ChemSensor* some certain predictions have reached 90% reliability. That is the case of *bellota* and *cebo intensivo* types.

Predictions carried out for *recebo*-type animals provided a reliable 81% whereas the *cebo extensivo*-type forecast is only 22%. This one is explained on the basis of the small number of samples used: a single failure over a small set gives necessarily a low percentage.

Compared to other technologies ChemSensor shows a clear advantage in terms of reliability, because it is based on gas chromatography and mass determination, and thus provides up to 500 variables per sample which are easily treated with *Pirouette* chemometrics software. This guarantees a reproducible analysis.

In addition, this technique does not use sensors, which are subjected to a limited lasting or aging. Chemsensor is not affected by environmental humidity and temperature conditions, so results gain accuracy and reproducibility.

The analysis time of each sample is very short, and may use the fatty acids of the sample or volatile compounds directly.

As used for the classification of dead animals is also perfectly suited for the analysis of the derived products such as a ham or loin. This technique is also a valuable tool in traceability tasks in industry.

Acknowledgments

Proyecto INIA ref: RTA2008-00026-C07-00.

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Genetic certification of the Iberian ham

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Abstract. Under current Spanish regulations, the pigs that provide the raw material for the preparation of the country's most appreciated meat-derived product, dry-cured Iberian ham, must be of a specific genetic composition. Only the Duroc breed is accepted for crossing with Iberian pigs, and a maximum of 50% of the Duroc genome is permitted in the animals used to make this ham. This study describes a set of statistical procedures for detecting the breed composition of Iberian ham via the use of multilocus genotypes obtained by the amplification of 25 microsatellite markers. The procedure proposed in this study has been used for several years routinely in our laboratory for various purposes such as to certificate the genetic composition of some Iberian registered trademark hams or to detect commercial fraud in the Iberian ham consumed in Spain.

Keywords. STR – Traceability – Individual assignment – Genetic admixture.

Certification génétique du jambon Ibérique

Résumé. *Les porcs qui produisent la matière première pour la préparation du jambon le plus populaire parmi les produits à base de viande de porc Ibérique doivent être d'une composition génétique spécifique selon la loi espagnole. Seule la race Duroc est acceptée pour le croisement avec le porc Ibérique, pour un maximum de 50% du génome, afin de produire ce jambon. Cette étude décrit un ensemble de procédures statistiques de détection de la composition génétique de la race porcine Ibérique à travers l'utilisation de génotypes multilocus obtenus par amplification de 25 marqueurs microsatellites. La procédure proposée dans la présente étude a été utilisée régulièrement pendant plusieurs années dans notre laboratoire à des fins diverses, comme la certification de la composition génétique de certaines marques de jambon Ibérique ou la détection des fraudes commerciales pour le jambon Ibérique consommé en Espagne.*

Mots-clés. *STR – Traçabilité individuelle – Brassage génétique.*

I – Introduction

The Iberian pig ham is one of the most important traditional food products of the Spanish culture. The added value of this breed was supported by the necessary conservation of the Mediterranean paddock, known as "dehesa".

Iberian ham quality is the result of the genetic composition, the animal feeding during the fattening and the artisan management, but to profite this traditional product we have to take into account important additional factors, as the marketing and commercialization.

We have developed a microsatellites based methodology to supervise the assignment of the individuals to the Iberian breed and its varieties, in order to support the conventional mechanism of supervising which is based on the intervention of a qualifier office that use the subjective observation of a morphological phenotype as criterion of assignment. Our methodology is based in the fitness of the genetic individual profile to population genetic profiles, previously defined in animals belonging to the breed herd book or admitted by the breeders association as integrated in the different varieties forming the Iberian breed group.

In the present paper we are showing a real experience using this methodology in the breed traceability of the products commercialized by Maldonado S.L. enterprise. It could be as a useful model to the sector to protect the genetic patrimony of the Iberian ham at the consumer eyes, the credibility.

II – Materials and methods

Maldonado S.L. is a traditional enterprise which commercialize only high quality Iberian products. Its “star” product is the called “Albarragena Ham” which is a selection of 100 hams among the thousands produced by the enterprise. This ham has been recognized as one of the most expensive ham of the world by the mass media.

These hundred hams have a lot of special commercial treatments to ensure their differentiated quality, such as a series number stamped in a silver medal, a presentation in an exclusive appearance, etc. This product reaches in the market the prize of 1500 Euro by piece.

One additional proof to support the credibility of the product is the incorporation of a certificate with the probability of assignment to the Iberian breed (Figure 1), and a series number to ensure the genetic traceability of the piece.

DNA extraction was performed from ham samples using the Genomic DNA Purification Kit (Gentra Systems, Minnesota, USA). Samples were screened for 25 pig microsatellites selected from the 27 markers recommended by the FAO for pig biodiversity studies (FAO,1998). Technical procedures of genotyping were described in previous papers (Vega-Pla *et al*., 2003; Garcia *et al*., 2006). Probability of assignment of individual genetic were determined using the Bayesian algorithms proposed by Baudouin *et al*. (2004), using Geneclass2 software (Piry *et al*., 2004) for the calculations. This methodology developed in the context of the EU project Characterization of genetic variation in the European pig to facilitate the maintenance and exploitation of biodiversity (BIO4 CT98-0188, DG XII European Commission, 1998-2000). It gave us the opportunity to access to a wide base of samples from almost all the pig European resources.

III – Results and discussion

All people involved in the Iberian pig world desire to maintain the credibility on the Iberian ham. There is an interest by the most serious private enterprises, which demanded an objective certification of purity of their products. Our methodology gave successfully response to this demand. Figure 1 shows a scheme of the factorial analysis results, defining the genetic assignment of the Albarragena hams to the Iberian pig genetic profile.

Table 1 is showing an example of the assignment probability of a set of Albarragena hams. This table of results is sent to the enterprise, to support its internal management in regards the selection of animals and pieces; the following up of the donor farms, etc.

Today the Iberian pig is researched intensely, also with the most advanced markers such as SNPs (Padilla *et al*. 2010), but these methodologies, under our point of view are optimal for marker assisted selection, and in the future for genomic selection, but microsatellite typing will continue being the tool of election for all around genetic characterization, breed traceability, and genetic diversity studies in general.

The present paper demonstrated that a tool to ensure the breed traceability of the Iberian pig products, also ensures the credibility of the high quality products.

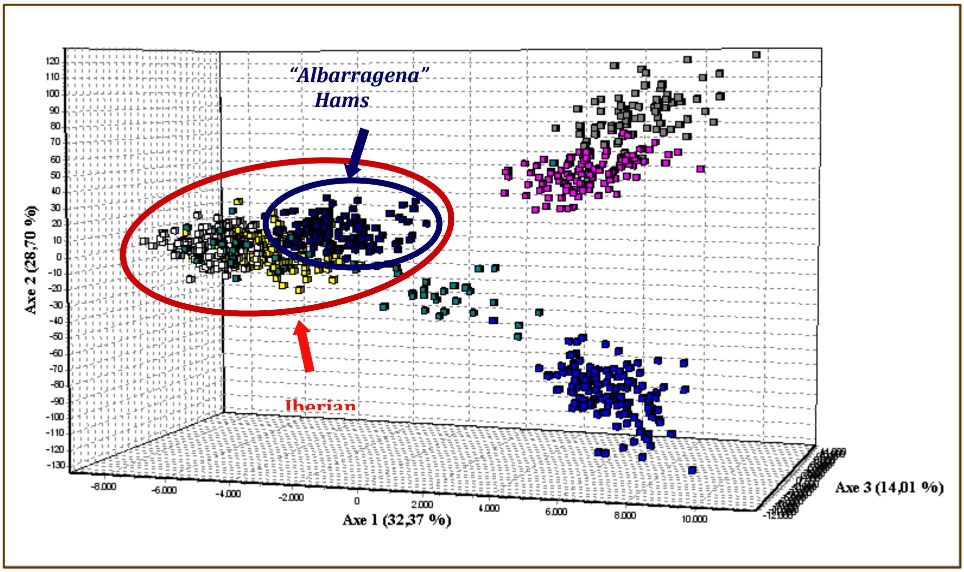

Fig. 1. **Factorial analysis results, showing the inclusion of the Albarragena hams in the Iberian genetic profile.**

Table 1. An example of the last Albarragena hams certified using the methodology of individual assignment to a population

Ham	P ASIG	Ham	P ASIG	Ham	P ASIG	Ham	P ASIG
0004393	0.983	0004893	0.991	0004839	0.986	0005087	0.992
0004663	0.944	0004878	0.993	0004454	0.988	0004217	0.991
0009386	0.990	0009287	0.923	0005626	0.984	0004010	0.987
0004842	0.991	0004362	0.969	0005118	0.990	0004369	0.967
0009550	0.991	0009435	0.985	0005047	0.993	0009378	0.992
0004344	0.940	0003560	0.969	0004455	0.981	0009392	0.974
0004970	0.989	0005235	0.990	0004329	0.951	0009321	0.990
0004461	0.984	0009454	0.978	0005233	0.987	0003618	0.991
0004871	0.992	0005050	0.984	0009343	0.979	0009445	0.994
0004509	0.972	0003557	0.967	0004951	0.990	0005245	0.993
0005025	0.985	0003930	0.992	0009335	0.961	0003621	0.986
0005259	0.988	0009295	0.949	0003568	0.994	0003492	0.994
0004478	0.984	0005169	0.984	0003652	0.983	0004861	0.993
0005241	0.977	0009437	0.966	0003573	0.973	0004034	0.905
0009545	0.990	0005044	0.996	0005052	0.985	0005084	0.986
0009460	0.956	0003707	0.987	0004915	0.994	0003623	0.966
0004375	0.962	0005177	0.994	0005035	0.990	0004095	0.994
0004920	0.987	0003939	0.992	0004504	0.981	0003650	0.954

IV – Conclusions

Today, Maldonado S.L. enterprise has enclosed in internal requirements of quality the use of the individual assessment bases on the genetic profiles obtained with a set of microsatellites, as an objective method to ensure the breed traceability of their Iberian pig products. This common applications is an example to follow to maintain n the credibility of the consumers.

Acknowledgements

The research was partially supported by the Project RZ00-15 of the Programa Nacional de Recursos y Tecnologías Agroalimentarias of Ministerio de Ciencia y Tecnología of Spain and the Consejería de Sanidad y Consumo de la Comunidad de Madrid, Spain

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Profil en acides gras dans différentes zones de jambons séchés et leur utilisation possible dans le cadre de la prévision de la qualité

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Résumé. Malgré le fait que les méthodes de classement selon la qualité de la carcasse des porcs Ibériques soient basées sur le prélèvement d'échantillons sur des animaux vivants ou récemment abattus, il est intéressant, au niveau commercial, de mettre au point de nouvelles méthodes en utilisant des échantillons de produits séchés. Dans cet article, nous présentons les résultats des analyses d'acides gras faites sur des échantillons de graisse de différentes parties de pièces séchées et le classement des pièces conformément à leur qualité. Sur 65 jambons, dont la génétique et l'alimentation sont connues, nous avons déterminé le profil en acides gras pour le lard prélevé sur 6 zones différentes afin de ne pas porter préjudice, ou très peu, à l'intégrité des pièces. Les zones choisies ont été le sommet du "V", la "punta" et la "maza", d'abord dans leur partie externe (VE, PE et ME) et ultérieurement celles qui correspondent aux 3 zones, mais plus à l'intérieur (VI, PI et MI). Il n'existe aucune différence pour les acides gras entre les zones internes et les zones externes. Ainsi, à partir de la 35ème pièce, ce sont les zones externes que nous avons analysées, ce qui ne détériore pas les jambons. Le pourcentage oléique est plus élevé de façon significative dans la VE, suivi de la ME et ensuite la PE, en considérant l'ensemble de toutes les pièces, ainsi que dans un groupe alimenté à base de glands. Ce n'est pas le cas chez les animaux élevés au fourrage, chez lesquels il n'y a pas de différences entre les zones. Les résultats de l'analyse discriminante révèlent que le pourcentage des assignations correctes est élevé et pour l'ensemble de toutes les pièces, il varie de 79,4% avec les deux AG de la PE, à 72,6% avec ceux de la ME.

Mot-clés. Porc Ibérique – Acides gras – Analyse discriminante.

Profile of fatty acids in distinct areas of cured hams and their possible use as quality indicators

Abstract. *Despite quality control methods in Iberian pigs being based on the collecting of samples from live or recently slaughtered animals, it is of commercial interest to fine tune other methods using samples from cured products. In this report the results of fatty acid analyses from fat samples from different parts of cured pieces of meat and the classification of the meats according to their quality will be presented. For sixty five hams, of known genetics and diet, the profile of fatty acids of pork fat extracted in six different places has been determined, chosen so that in none or very few cases the integrity of the piece of meat would be affected. The chosen places were in the vertex of the "V", the "punta" (the thigh end of the ham) and the "maza" (the meatiest part of the ham), first in its external part (VE, PE and ME respectively) and then the three interior areas corresponding to the aforesaid areas (VI, PI and MI). There are no differences between the fatty acids in the internal areas and the external areas. Thus, from ham number thirty five upwards samples were only taken from the external areas, which didn't deteriorate the hams. The percentage of oleic acid is significantly higher in the VE samples, followed by ME and then by PE, considering the group of hams as a whole and those bred free-range. This doesn't happen in factory farmed animals, in which there are no differences between the areas. The results of the differentiated analysis show that the percentage of correct assignments is elevated and in the group containing all of the hams it varies from 79.4% with the AG to PE samples to 72.6% with the ME samples.*

Keywords. *Iberian pork – Fatty acids – Discriminate analysis.*

I – Introduction

Dans l'actualité, toutes les méthodes pour vérifier la qualité, en ce qui concerne l'alimentation du porc Ibérique, retombent exclusivement sur le premier échelon du secteur, c'est-à-dire, l'éleveur. Rien n'a été néanmoins réalisé au niveau suivant, étant celui du secteur industriel. L'objectif de ce travail est d'analyser l'influence de l'alimentation de l'animal qui donne le jambon séché et qui a subi des régimes alimentaires différents.

II – Matériel et méthodes

Soixante-cinq jambons ont été utilisés. Ils ont été fournis par diverses industries du secteur et ils correspondent à 7 catégories selon la génétique (100% Ibérique pur, croisé à 50% avec Duroc Jersey, et 0% de sang Ibérique) et le type d'alimentation dans l'étape finale d'engraissement ("Bellota", "Recebo" y "Cebo").

Des échantillons de tissus adipeux de 3 zones différentes de chaque jambon ont été prélevés de manière à ne pas altérer le moins possible le produit commercial. Les zones sélectionnées ont été: de la base du "V", de la Masse et de la Pointe du jambon (Fig. 1). Dans le cas de 30 jambons, les couches externe et interne des échantillons de tissus adipeux prélevés ont été séparées afin de déterminer dans chacune d'elles la composition en tissus adipeux. Dans le reste des échantillons, les deux couches ont été analysées ensemble. Les analyses ont été réalisées au Laboratoire Agroalimentaire de la Junta de Andalucía à Cordoue, selon la normative officielle d'analyses.

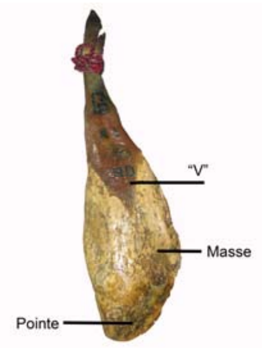

Fig. 1. Zones de prélèvement des échantillons sur le jambon.

Le pack de programmes statistique SPSS a été utilisé pour l'analyse statistique, comparant les valeurs obtenues dans chaque groupe moyennant un test Ducan, et la caractérisation de chaque groupe, en fonction de leur composition en acides gras, a été réalisée grâce à une analyse discriminante.

III – Résultats et conclusions

La composition en acides gras des couches de tissu adipeux des jambons, de chaque zone, est recueillie dans le Tableau 1.

Dans le Tableau 2, on peut voir les valeurs (moyenne, maximale et minimale), du contenu en acide oléique de la couche externe, de chacune des zones du jambon, selon le type d'alimentation des animaux. On peut y apprécier que les valeurs les plus élevées en acide oléique sont obtenues dans la couche externe du "V" et les minimales dans la Pointe.

Tableau 1. Composition des couches de tissu adipeux des jambons de porc Ibérique (n=30)

Acides gras	Couche	Masse		Pointe		V	
		Moyenne	Signif.	Moyenne	Signif.	Moyenne	Signif.
Palmitique	Externe	23.2	0.69	24.6	0.01	22.2	0.89
	Interne	23.1		23.5		22.3	
Stearique	Externe	12.3	0.93	13.8	0.20	11.0	0.95
	Interne	12.3		13.2		11.0	
Oleique	Externe	50.4	0.51	48.4	0.49	53.1	0.78
	Interne	49.9		48.9		52.9	
Linoleique	Externe	7.8	0.03	7.2	0.00	6.9	0.12
	Interne	8.5		8.2		7.5	

Tableau 2. Contenu en acide oléique de la couche externe du tissu adipeux de jambons séchés selon le type d'alimentation des animaux

Tipe de jambon	Zone	N	Moyenne ± D.e.	Mínim.	Máxim.
Bellota 100	ME	15	53,2 ± 1,8 [b]	50	56
	PE	16	51,7 ± 2,3 [a]	50	58
	VE	16	54,9 ± 2,3 [c]	51	58
Bellota 50	ME	5	51,6 ± 2,1 [a]	49	54
	PE	6	50,1 ± 2,2 [a]	46	52
	VE	6	56,4 ± 1,0 [b]	55	58
Recebo 100	ME	13	51,7 ± 2,0 [b]	47	54
	PE	12	49,9 ± 2,5 [a]	46	53
	VE	12	53,3 ± 2,5 [c]	48	57
Recebo 50	ME	3	51,1 ± 2,4 [a]	49	53
	PE	3	47,8 ± 3,1 [a]	44	50
	VE	3	55,1 ± 0,1 [b]	55	55
Cebo 100	ME	15	48,4 ± 2,3 [b]	46	53
	PE	15	46,7 ± 2,5 [a]	43	53
	VE	15	51,5 ± 2,6 [c]	47	57
Cebo 50	ME	8	48,6 ± 1,5 [b]	47	51
	PE	8	46,3 ± 2,5 [a]	43	51
	VE	7	49,9 ± 2,4 [b]	47	53
Cebo 0%	ME	3	47,0 ± 1,9 [b]	46	49
	PE	3	43,7 ± 1,9 [a]	42	46
	VE	3	46,4 ± 2,1 [ab]	44	49
Toutes	ME	62	50,6 ± 2,8 [b]	45,5	56,1
	PE	63	48,8 ± 3,3 [a]	42,3	58,3
	VE	62	53,0 ± 3,3 [c]	44,4	58,1

ME: Masse Externe; PE: Pointe Externe; VE: V Externe.

Les résultats de l'application de l'analyse discriminante au total des jambons sont recueillis dans les Tableaux 3, 4 et 5. Dans ces tableaux on peut apprécier que le pourcentage le plus élevé des échantillons correctement classées est obtenu en utilisant la composition des acides gras de la graisse de la couche externe de la pointe (77,8%). Dans la catégorie intermédiaire "Recebo" le pourcentage d'erreurs de classement obtenu a été plus élevé, si l'on considère que ceci est dû à la grande hétérogénéité des animaux.

Tableau 3. Résultats du classement des jambons selon la composition en acides gras de la graisse externe de la Masse

Type commercial	N	B100	B50	R100	R50	P100	P50	P0
B100	15	73,3	0,0	13,3	0,0	6,7	6,7	0,0
B50	5	0,0	100,0	0,0	0,0	0,0	0,0	0,0
R100	13	23,1	0,0	61,5	0,0	7,7	7,7	0,0
R50	3	0,0	0,0	0,0	100,0	0,0	0,0	0,0
P100	15	0,0	0,0	6,7	0,0	73,3	6,7	13,3
P50	8	0,0	0,0	12,5	0,0	0,0	87,5	0,0
P0	3	0,0	0,0	0,0	0,0	33,3	0,0	66,7

B: Bellota; R: Recebo; C: Cebo; Classés correctement 77,8 % des échantillons.

Tableau 4. Résultats du classement des jambons selon la composition en acides gras de la graisse externe de la Pointe

Type commercial	N	B100	B50	R100	R50	P100	P50	P0
B100	15	87,5	0,0	12,5	0,0	0,0	0,0	0,0
B50	5	0,0	100,0	0,0	0,0	0,0	0,0	0,0
R100	13	25,0	0,0	47,7	0,0	16,7	16,7	0,0
R50	3	0,0	0,0	0,0	100,0	0,0	0,0	0,0
P100	15	6,7	0,0	0,0	0,0	80,0	13,3	0,0
P50	8	0,0	0,0	12,5	0,0	12,5	75,0	0,0
P0	3	0,0	0,0	0,0	0,0	0,0	0,0	100,0

B: Bellota; R: Recebo; C: Cebo; Classés correctement 77,8 % des échantillons.

Tableau 5. Résultats du classement des jambons selon la composition en acides gras de la graisse externe du V

Type commercial	N	B100	B50	R100	R50	P100	P50	P0
B100	16	68,8	6,3	12,5	6,3	6,3	0,0	0,0
B50	6	0,0	83,3	0,0	16,7	0,0	0,0	0,0
R100	12	16,7	8,3	50,0	8,3	8,3	0,0	8,3
R50	3	0,0	0,0	0,0	100,0	0,0	0,0	0,0
P100	15	0,0	0,0	6,7	0,0	86,7	0,0	6,7
P50	7	0,0	0,0	14,3	0,0	14,3	28,6	42,9
P0	3	0	0	0	0	0	0	100

B: Bellota; R: Recebo; C: Cebo; Classés correctement 69.4 des échantillons.

Cette méthode d'analyse peut donc être un outil utile pour le classement des jambons séchés et avec un pourcentage d'erreur comparable à ceux qui se produisent quand on évalue l'alimentation des animaux à travers la composition en acides gras d'échantillons de biopsies du tissu adipeux sous cutané. Ces erreurs sont finalement la conséquence de la fragilité du système basé sur le trait des acides gras pour la détermination de la qualité (Porras et al., 2009), aussi bien dans les biopsies que dans les échantillons de tissu adipeux des propres jambons. La nouveauté de ce système, une fois mis au point, visant à fixer les intervalles des principaux acides gras, permettrait de contrôler la qualité au niveau du séchoir et pas seulement au niveau de l'exploitation, comme on le fait actuellement.

Remerciements

À la Junta de Andalucía et à INIA, qui ont financé les projets de recherche PIA4.01.3 et CAL02-025-C4-2, respectivement.

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Utilization of tocopherols quantification for the differentiation of Iberian pigs fed in free-range or with feeds in intensive or extensive systems in different geographical regions

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Abstract. The aim of this work was to study the capacity of differentiation of the technique based on the tocopherols quantification in blind samples of fat and muscle coming from animals fed (1) with feed in an intensive system, (2) with an enriched-fat diet in extensive conditions, (3) with a conventional diet in extensive conditions, (4) recebo (low fattening in free-range) and (5) exclusively free-range in different geographical areas. The technique was able to differentiate ($P < 0.05$) samples from animals fed in free-range from the other groups. It was also able to classify ($P < 0.05$) by muscle gamma-tocopherol quantification, animals fed with feed in intensive situations from those fed in recebo or with feed in an extensive system. However, there were not significant differences in the content of gamma-tocopherol in fat and muscle between the recebo pigs (with average weight gained free-range <25 kg) and those fed with feed in extensive conditions.

Keywords. Iberian pig – Tocopherols – Carcass classification – Quality.

Utilisation de la détermination du tocophérol pour la différenciation de porcs Ibériques alimentés au pâturage ou avec du concentré dans les systèmes d'exploitation intensive ou extensive dans différentes régions géographiques

Résumé. *L'objet de ce travail est d'étudier la capacité de différenciation de la technique basée sur la détermination des tocophérols que contiennent les échantillons en aveugle de graisses et de muscles en provenance d'animaux engraissés dans des systèmes (1) intensifs, (2) extensifs et alimentés à base de concentrés contenant de la graisse, (3) en engraissement extensif à base de concentrés conventionnels, (4) ré-engraissement, et (5) élevés au pâturage dans différentes régions géographiques. La technique a été capable de différencier (P<0.05) les échantillons provenant d'animaux alimentés au pâturage par rapport au reste du groupe. On a également pu classifier (P<0.05) les animaux alimentés à base de concentrés en engraissement intensif par rapport à ceux qui ont été alimentés dans les exploitations de ré-engraissement ou engraissement extensif. Toutefois, il n'y a pas eu de différences significatives dans les teneurs en gamma-tocophérol entre les animaux de ré-engraissement et ceux d'engraissement en exploitation extensive.*

Mots-clés. *Porcs Ibériques – Tocophérol – Classification carcasse – Qualité.*

I – Introduction

The high price that the Iberian pig products reaches in the market has originated faked practices in order to marketing products coming from Iberian pigs fed with feeds in confinement as if they were from pigs fed free-range with acorns and grass. As quality control measure during the last years has been used the profile of fatty acids to determine the productive origin of the animals, however this technique has stopped to be an approach of classification of full validity. Among the alternative techniques of classification is tocopherols determination. This measurement is interesting since tocopherols, which are found at high concentrations in acorns (mainly gamma-tocopherol) and grass (mainly alpha-tocopherol), are accumulated in fat tissues (Rey *et al*., 1997). This accumulation has also been found to depend on the days that pigs stayed in free-range conditions or the kilograms of fattening and so could discriminate the feeding background of the animals and its products classification (Rey *et al*., 2006). Previous studies (Rey *et al*., 1997, Daza *et al*., 2005, Rey *et al*., 2006) reported differences in the tocopherol concentration between groups fed free-range and those fed in intensive conditions with mixed diets. However, no information is available on the tocopherols accumulation with other feedings such as enriched-fat or conventional diets in combination with extensive conditions in which pigs could utilizes some natural resources (mainly grass) (label recently defined in the Iberian pig Quality Norm) (BOE, 2007) and its possible utilization to differentiate these groups from the others. The accumulation of tocopherols at different geographical locations because of grass availability has also to be explored. The aims of this work were to study the capacity of differentiation of the technique based on the tocopherols quantification in blind samples of fat and muscle from animals fed different feedings in intensive or extensive conditions.

II – Materials and methods

The experimental groups (Table 1) consist of Iberian pigs (pure Iberian or crossed) with ages between 10-18 months fed different dietary treatments.

Table 1. Data of the Iberian pigs from the different experimental groups

N	BREED	AGE (months)	FARM	SITUATION	FEEDING	DAYS FREE-RANGE	Fattening free-range (kg)	Grass resources
13	Pure Iberian pigs (males)	18	San Amaro	Olivenza (Badajoz)	Enriched fat diet (extensive)	0	0,00	low
32	Crossed 50 %	10	Turcañada	Fuenteovejuna (Córdoba)	Conventional mixed diet (intensive)	0	0,00	
12	Pure Iberian pigs Valdesequera	12	Valdesequera	Badajoz	Free-range	60	40,15	medium
32	Crossed 75 %	14	Puerto Lobo	Cabeza la Vaca (Sur Badajoz)	Free-range	72	58,39	high
10	Pure Iberian pigs Valdesequera	12	Valdesequera	Badajoz	Conventional mixed diet (extensive)	0	0,00	medium
12	Pure Iberian pigs Valdesequera	12	Valdesequera	Badajoz	Recebo	46	17,13	medium
6	Pure Iberian pigs Valdesequera	12	Valdesequera	Badajoz	Conventional mixed diet (extensive)	0	0,00	medium
14	Pure Iberian pigs Valdesequera	12	Valdesequera	Badajoz	Recebo	46	18,07	medium
29	Pure Iberian pigs (Castrated females)	12	San Miguel	Ciudad Rodrigo (Salamanca)	Free-range	91	55,22	high
7	Pure Iberian pigs Valdesequera	12	Valdesequera	Badajoz	Conventional mixed diet (extensive)	0	0,00	medium
13	Pure Iberian pigs Valdesequera	12	Valdesequera	Badajoz	Recebo	46	23,08	medium

One group was fed with a commercial feed in confinement, a second group was fed outdoors (limited pens with access to grass) with an enriched-fat diet which contained (g/kg) 268 g grease-out wheat meal, 268 g grease-out barley meal, 100 g grease-out corn meal, 150 carob-bean meal, 150 g sunflower seed, 50 g pea, 3 g calcium carbonate, 6 g dicalcium phosphate, 4 g sodium chloride. A third group was fed outdoors (similar conditions that second group) with a conventional diet that contained (g/kg) 120 g corn, 111 g wheat, 110 g barley, 98 g soya-bean meal, 37 g DDG barley, 15 g sunflower seed, 10 g beet root pulp, 7 g dicalcium phosphate, 4 g pork lard, 4 g calcium carbonate, 2 g sodium chloride. Another three experimental groups were fed in “recebo” (free-range for a period of 46 days and then received the same conventional mixed diet that those pigs fed in extensive conditions) and finally another three groups were fed free-range during a variable period. Pigs fed free-range were from different locations in Spain and hence they had variable grass availability (medium and high). Also pigs fed in extensive conditions had different grass resources depending on the pens location.

Pigs were slaughter at the average weight of 164,3 kg (±16,4) and samples (fat at the level of the tail and muscle from different areas) were collected and frozen at -20°C until analysis. Tocopherols (gamma and alpha) in muscle were quantified according to the method described by Rey *et al*. (1996) in which samples were homogenised in dibasic sodium phosphate buffer and extraction was made with ethanol and hexane. Tocopherols in fat were analysed by saponification in presence of pyrogalol, potassium chloride and potassium hydroxide as described before (Rey *et al*., 2006). In both methods tocopherols were dissolved in ethanol prior to analysis by reverse-phase HPLC (HP 1100, with a diodo array detector) (Hewlett Packard, Waldbronn, Germany). Separation of gamma and alpha tocopherols was made by RP-C18 column at a flow rate of 2ml/min. (methanol: water 97:3).

Data were analysed using the general linear model (GLM) procedure contained in SAS version 8 (SAS, 1999). The comparative analysis between means was conducted using the Duncan's test.

Groups of free-range pigs had concentrations of gamma-tocopherol in fat (ug/g: 0.77, 1.51 and 1.80) according to the days in freedom (60, 77 and 91), the weight gained (40, 58 and 55 kg) and the grass resources (medium, high and high, respectively). Alpha-tocopherol was also affected by these factors (ug/g: 8.3, 11.4 and 10.8). In those pigs ("recebo") that stayed in freedom for a short period (46 days) and so gained less weight (17, 18 and 23 kg) the concentration of tocopherols in fat were lower as expected (gamma-tocopherol ug/g: 0.31, 0.26 and 0.58; alpha-tocopherol ug/g: 6.3, 6.6 and 9.5 respectively). In muscle, due to the heterogeneity of the sample and so the different intramuscular fat content differences were not as marked as in fat.

On the other hand, groups fed with a conventional diet outdoors in delimited pens had variable concentrations of tocopherols in fat (gamma ug/g: 0.38, 0.34 and 0.52; alpha ug/g: 6.5, 6.3 and 8.9) and muscle (gamma ug/g: 0.18, 0.13 and 0.15; alpha ug/g: 2.6, 1.9 and 2.5) probably in function of the different natural resources available.

Fat samples (n=180) were classified by 74 % correctly. The main mistake was in the classification of pigs fed outdoors in delimited pens that were considered as those fed extensively in freedom for a short period ("recebo"). Classification of muscle samples (n=188) was correct by 72 % even though the high variability in the sample collection and showed the same trend observed in fat.

Average results of tocopherols concentration in fat and muscle from the experimental groups are presented in Tables 2 and 3. In fat (Table 2), gamma-tocopherol concentration was higher (P=0.0001) in free-range pigs than the other groups. Free-range pigs had also higher concentrations of alpha-tocopherol than the other groups with the exception of those pigs that received an enriched-fat diet, which had similar concentrations. The alpha-tocopherol concentration was also of interest to discriminate between pigs fed with a conventional mixed diet in intensive conditions and the other groups. These pigs fed indoors with feed had the lowest alpha-tocopherol concentration (P=0.0001). Neither gamma-tocopherol nor alpha-tocopherol was different in fat from those pigs fed extensively in freedom ("recebo") or in pens with a conventional mixed diet.

Table 2. Tocopherols (gamma and alpha) concentration (ug/g) in fat from the experimental groups (average values between similar groups) according to their feeding

Conditions	**Extensive in freedom**		**Extensive in delimited pens**		**Intensive**	**RMSE**	**P**
Feeding	**Free-range**	**Recebo**	**CMD†**	**EFD†**	**CMD**		
Gamma-tocopherol (ug/g)	1.501 a	0.381 b	0.415 b	0.351 b	0.367 b	0.37	0.0001
Alpha-tocopherol (ug/g)	10.653 a	7.490 b	7.216 b	10.045 a	4.364 c	1.97	0.0001
Gamma/Alpha (ug/g)	0.142 a	0.050 c	0.058 c	0.036 c	0.088 b	0.04	0.0001

†CMD: Conventional mixed diet; EFD: Enriched-fat diet.

Table 3. Tocopherols (gamma and alpha) concentration (ug/g) of muscle samples from the experimental groups (average values between similar groups) according to their feeding

Conditions	**Extensive in freedom**		**Extensive in delimited pens**		**Intensive**		
Feeding	**Free-range**	**Recebo**	**CMD†**	**EFD†**	**CMD**	**RMSE**	**P**
Gamma-tocopherol (ug/g)	0.476 a	0.153 b	0.150 b	0.137 b	0.0886 c	0.09	0.0001
Alpha-tocopherol (ug/g)	3.925 a	2.948 b	2.341 c	4.022 a	1.7553 d	0.80	0.0001
Gamma/Alpha (ug/g)	0.124 a	0.0546 c	0.066 b	0.037 d	0.0522 c	0.02	0.0001

†CMD: Conventional mixed diet; EFD: Enriched-fat diet.

In muscle, differences between groups were more marked than in fat (Table 3). Gamma-tocopherol concentration was statistically different in those pigs fed in intensive conditions and in free-range in comparison with the other groups fed in extensive conditions either in limited pens or recebo (P=0.0001). In muscle as in fat, alpha-tocopherol was the lowest in those pigs fed in intensive conditions while the highest concentrations were detected in both enriched-fat diet in extensive conditions and in free-range groups. Alpha-tocopherol quantification in muscle also allowed the discrimination between pigs fed recebo than those fed a conventional diet in extensive conditions. Hence, those pigs fed recebo showed higher alpha-tocopherol concentrations than those fed a conventional diet extensively (2.9 *vs* 2.3).

III – Conclusions

The determination of gamma-tocopherol in fat or muscle from Iberian pig allows the discrimination of free-range pigs from others fed in intensive or in extensive conditions with conventional or enriched-fat diets. Gamma-tocopherol quantification also allows a clear differentiation between free-range pigs and those that stayed a short period and gained low weight (<20-25 kg) in free-range ("recebo"). However, the quantification of both tocopherols (alpha and gamma) in different tissues is needed to differentiate pigs fed mixed diets in extensive or intensive conditions between them and from the other groups.

Acknowledgements

The authors wish to acknowledge the financial support received from the INIA (RTA2008-00026).

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Volatile compounds and sensorial traits of "Toscano" dry-cured ham

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Abstract. The aims of this study were to evaluate the effect of seasoning time on aromatic profiles and to describe the sensorial profile of "Toscano" dry-cured ham. At this aim ten hams, cured according to the rule of "Toscano" PDO label, were sampled at the begin of seasoning and at 1, 3, 6 and 12 months. The samples of muscular tissue were submitted to volatile compounds analysis by SPME-GC/MS. At the end of seasoning a sensorial analysis of hams was carried out by a trained panellist group. As regard volatile analysis, about 80 compounds were identified belonging to: aldehydes, organic acids, ketones, esters, alcohols and furans. Aldehydes represent the most important chemical family in "Toscano" ham; they showed a significant increase only in the first 6 months of seasoning. The second quantitatively important chemical family in "Toscano" ham was that of the organic acids that increased along the whole curing time. Ketones dramatically decreased at 12 months of seasoning while esters showed a more constant trend. Significant correlations were found among sensorial traits and aldehydes compounds.

Keywords. Volatile analysis – Sensorial profile – Dry-cured ham – Seasoning.

Les composés volatils et les caractéristiques sensorielles du jambon sec "Toscano"

Résumé. *Les objectifs de cette étude étaient d'évaluer l'effet du temps de mûrissage sur les profils aromatiques et de décrire le profil sensoriel du jambon sec "Toscano". Dix jambons mûris selon la règle du label AOP "Toscano", ont été échantillonnés au début du mûrissage et à 1, 3, 6 et 12 mois. Les échantillons de tissu musculaire ont été soumis à l'analyse des composés volatils par SPME-GC/MS. À la fin du mûrissage l'analyse sensorielle des jambons a été réalisée par un groupe d'experts qualifiés. Pour ce qui concerne l'analyse des composés volatils, environ 80 composés ont été identifiés appartenant à: aldéhydes, acides organiques, cétones, esters, alcools et furanes. Les aldéhydes représentent la famille la plus importante dans le jambon "Toscano", et ils ont montré une augmentation significative seulement dans les 6 premiers mois de mûrissage. La seconde famille de substances chimiques la plus importante quantitativement dans le jambon "Toscano" a été celle des acides organiques qui ont augmenté pendant tout le temps de mûrissage. Les cétones ont diminué considérablement à 12 mois de maturation tandis que les esters ont montré une tendance plus constante. Des corrélations significatives ont été trouvées parmi les caractères sensoriels et les composés aldéhydes.*

Mots-clés. *Analyse de composés volatils – Profil sensoriel – Jambon sec – Mûrissage.*

I – Introduction

Several studies have been dealing with the flavour of ham of Mediterranean area (Dirinck *et al*., 1997), either on the relationships between compositional traits and sensory qualities of French and Italian dry-cured ham or on the volatile profile of Iberian hams and on its evolution with ripening, with the identification of the compounds primarily responsible for the typical aromatic characteristics. Also, Italian Parma and San Daniele hams have been characterized for their aromatic composition (Careri *et al*., 1993, Bolzoni *et al*., 1996; Gaspardo *et al*., 2008).

Conversely, the information on the ""Toscano"" dry-cured ham are, at present, limited to physical-chemical traits as affected by genetic type and rearing system (Franci *et al*., 1997; Pugliese *et al*., 2005; Pugliese *et al*., 2010), while information on its volatile composition are still lacking. There are also few details on its sensory properties. So the aim of this trial is to investigated the evolution of volatile compounds profile of "Toscano" dry-cured ham during 12 months ripening by SPME-GC-MS and, at the same time, to describe its sensorial profile at the end of the 12 months of seasoning.

II – Materials and methods

Ten hams were seasoned for 12 months according to the "Toscano" PDO Consortium ripening protocol and *Biceps Femoris* muscle was sampled immediately after trimming and after 1, 3, 6, 12 months of ripening. The volatile compound profile was obtained by SPME–GC-MS technique. An Agilent 7890 GC-Chromatograph equipped with a 5975A MSD with EI ionization was used for analysis. Volatile compounds were identified by matching EI mass spectra against NIST 05 or Wiley 07 spectral library and Kovats indices. As regard sensorial analysis samples of cured ham were assessed by a trained panel of 10 members using a descriptive analysis method. A 100 mm unstructured scale was used. Three-four hams were evaluated simultaneously in each evaluation session and the samples order was randomised. Data were subjected to analysis of variance using, as fixed effects, "ham" (10 levels) and "time of ripening" (5 levels) for evolution of aromatic profile or "panellist" (10 levels) for sensorial data. Significant differences were tested after Student's t test. Principal Component Analysis (PCA) was applied to evaluate the relationships among variables.

III – Results

Over 60 compounds belonging to esters, aldehydes, organic acids, ketones and alcohols were identified but for lack of space the means of each aromatic compounds were not tabulated while their evolution is shown in Fig. 1.

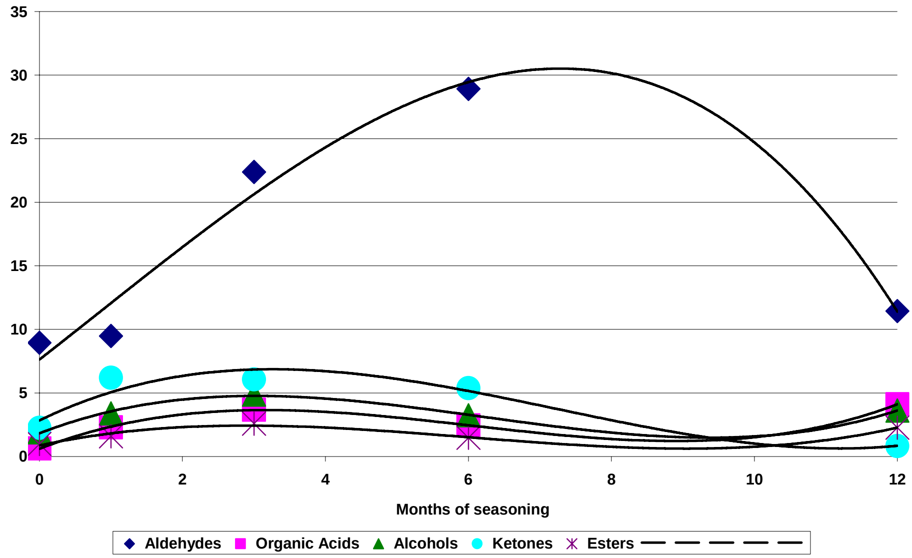

Fig. 1. Evolution of the main chemical families according to seasoning time.

Aldehydes were the most represented compounds in "Toscano" ham, similarly to what reported on lean tissue of Iberian cured ham (Ruiz *et al*., 1999). The aldehydes and ketones groups reached their greatest relative concentration at 6 and 1 month of ripening, respectively, and then decreased significantly, probably due to further reactions with other compounds. Alcohols, organic acids and esters concentration increased until the third month of ripening, and remained rather constant until 12 months. The described trend of the chemical families was strictly linked to the drying/ripening conditions when most of the biochemical changes become relevant. In the ripening stages a temperature of 16-18°C is adequate for the activity of the lypolitic and proteolytic enzymes, which progressively decrease thereafter, probably due to the contemporary water activity decrease along the process.

Table 1 shows the means and standard deviations of the sensory traits. The variability of the traits was quite high, that means that “Toscano” dry-cured ham is characterized by a medium-low degree of homogeneity. This can be considered a problem from commercial point of view but, on the other hand, it is a guarantee of a really traditional production process.

Table 1. Sensorial analysis of "Toscano" ham

Parameter	Means (mm)	Significance of "ham effect"	Standard deviation
Yellowness	33.10	<.0001	21.34
Pinkness	40.49	0.0056	18.08
Oiliness	41.90	0.0106	18.70
Firmness of fat	42.62	0.6815	17.76
Internal fat knob	40.39	0.0050	15.92
Quantity of external fat	41.33	0.1583	16.94
Redness	47.30	0.0005	16.26
Uniformity of color	42.83	0.0024	15.40
Marbling	44.07	<.0001	15.60
Firmness of lean	48.00	0.0015	15.72
Odor intensity	53.83	0.4942	16.67
Saltiness	35.48	0.1439	20.74
After taste	53.06	0.3729	15.84
Rancidity	7.25	0.8018	12.05
Hardness	30.69	0.4049	17.86
Juiciness	50.42	0.0004	22.63
Persistence	54.80	0.3566	19.99
Overall acceptability	56.79	0.0634	17.55

The variability of the data is high only for what concerns the data of color and of fat (quantity and quality). For other parameters, related in particular to aroma and odor, the variability among hams was very small. This fact attests to the validity of the curing process that can provide homogeneous products in these respects. As regards PCA, two principal components were generated that accounted for 60% of the total variance (PC1 = 39% and PC2 = 21%). The score plot of the samples is shown in Fig. 2. For PCA only aldehydes compounds were used because they were the only compounds that showed significant correlations with sensorial traits. As it can see in the figure the sensorial traits link to taste are very poor related with aldehydes compounds. The only positive relationships were found with 13-octadecenal, benzeneacetaldeyde, benzaldeyde and nonanal that seems to be related to rancid aroma (García-González *et al*., 2008).

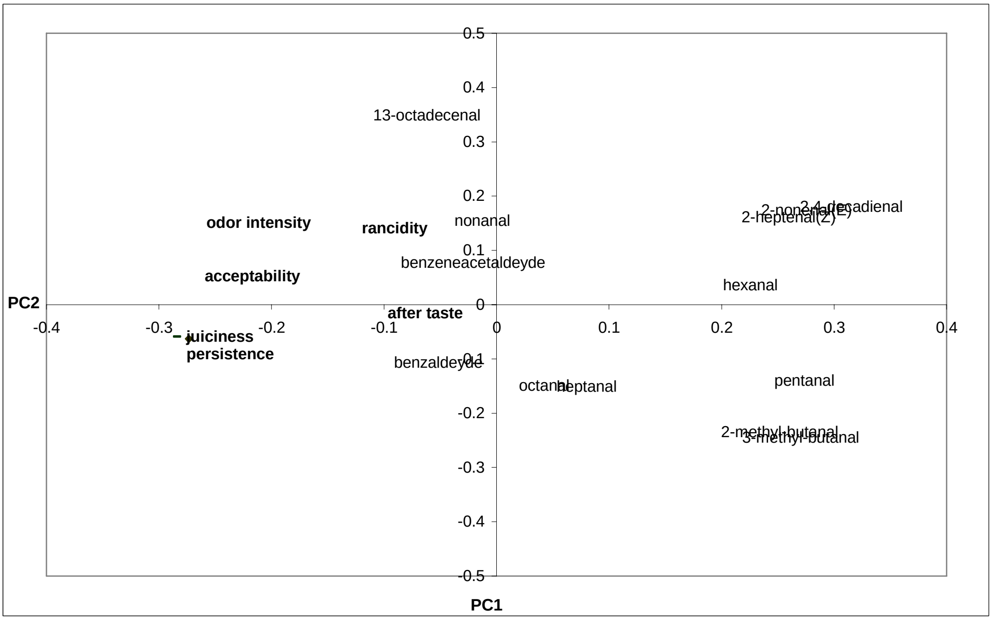

Fig. 2. Principal Component Analysis results.

Acknowledgements

The authors wish to acknowledge the Agenzia Regionale Toscana per lo Sviluppo e l'Innovazione nel settore Agricolo e forestale (ARSIA) for the financial support and Mr. Aldo Neri of the "Prosciuttificio Valdinievole" for the technical support.

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Non-destructive analysis of fresh Iberian pork loins by near-infrared spectroscopy (NIRS)

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Abstract. A fiber-optic contact probe near infrared spectroscopy (NIRS) instrument is evaluated to predict fat, moisture and protein in intact Iberian pork loins. A total of 173 Iberian pig loin samples were analyzed. Spectra from intact loins were collected with a LabSpec®Pro A108310 spectrometer (Analytical Spectral Device) and from the ground meat with a FossNIRSystem instrument. Spectra attenuation was required for removing very signal noise areas in the intact loin spectra. As reference values were used the NIRS predictions of the sample set estimated with a robust calibration model for ground samples and analyzed in a FossNIRSystem instrument. Modified Partial Least Squared (MPLS) regression was performed and different spectra pretreatments and spectral regions evaluated. The range 450-2300 nm performed the best models for the instrument evaluated, showing a suitable potential of the NIRS instrument for on-line analysis of pork loins.

Keywords. Iberian pigs – Loins – NIR Spectroscopy – Fiber optic – Fat – Protein – Moisture.

Analyse non destructive d'échines fraîches de porc Ibérique au moyen de spectrospie à infrarouge proche (NIRS)

Résumé. *Un équipement spectroscopique à infrarouge proche (NIRS) doté d'une sonde de fibre optique de contact est évalué pour la prédiction de la graisse, de l'humidité et de la protéine dans des échines intactes de porcs Ibériques. Un total de 173 échantillons d'échines de porcs Ibériques ont été analysés. Les spectres ont eu besoin d'être atténués pour éliminer des aires ayant un signal bruyant. Comme valeurs de référence on a utilisé les prédictions NIRS de l'ensemble des échantillons estimées avec un modèle robuste de calibrage créé pour des échantillons hachés et analysés par un instrument FossNIRSystem. L'algorithme de régression des Moindres Carrés Partiels Modifiés (MPLS) a été utilisé pour évaluer différents pré-traitements et régions spectrales. L'intervalle 450-2300 nm a fourni les meilleurs modèles pour l'instrument évalué, en montrant le potentiel d'analyse en ligne de l'instrument NIRS pour l'échine de porc.*

Mots-clés. *Porcs Ibériques – Rein – Spectroscopie NIR – Fibre optique – Matières grasses – Protéines – Humidité.*

I – Introduction

Iberian pig products have a high rate consumer acceptance, leading to high prices in the market. Consumers assess the quality of those products based on the exceptional organoleptic, healthy and sensory characteristics, but guarantee a target meat product is a complex task that requires control procedures (Prieto *et al.*, 2009a). Fresh meat is a very heterogeneous product and the determination of the major chemical constituents, such as fat, moisture or protein, is interesting for labelling purposes or for preparing good mixtures to produce dry-cured or processed products.

Traditional wet chemistry to determine these parameters are time-consuming, tedious, costly and destructive methods. Nowadays, the meat industry has shown a great interest for new technologies that enables fast, accurate, non-destructive quality analysis. Near-Infrared Spectroscopy (NIRS) has shown its potential to predict chemical composition in meat products (Prevolnik *et al.*, 2004; Prieto *et al.*, 2009a), even in on-line applications in the industry (Tøgersen *et al.*, 1999; González-Martín *et al*., 2002; Huang *et al*., 2008; Prieto *et al*., 2009b) resolving part of the demands of the industry. Most of the literature has used ground samples which require a sample preparation. In this study is evaluated a fiber-optic contact probe NIRS instrument to predict chemical composition (fat, moisture and protein) of intact Iberian pork loins.

II – Materials and methods

1. Sample set and NIRS measurements

One hundred and seventy three samples of Iberian pork loins were analyzed. The samples were taken from the beginning of the loin in the shoulder area of the animals. Two NIRS analyses were performed with different instruments and samples presentation.

A post-dispersive diode array scanning monochromator spectrometer LabSpec®Pro A108310 (Analytical Spectral Device-ASD Inc., Boulder, Colorado, USA) working from 350-2500 nm (1nm spectral resolution) in reflectance mode was used to analyze intact pork loin samples at the slaughter house, two hours post-mortem. The loins after being analyzed with the LabSpec®Pro were vacuum packaging and frozen at -20ºC to be stored. A FossNIRSystem 6500 equipped with a spinning module for standard circular cups working from 400-2500 nm (2 nm spectral resolution) in reflectance mode was used to analyze the same samples set in ground presentation. Before recording the NIRS spectra, the muscles were ground and homogenized by a vertical cutter mixer (Heidolp homogenizer DIAX 900). Two spectra per sample were measured in each case.

2. Data modelling

Chemometric data treatment was performed using the software WinISI II ver 1.50 (Infrasoft International, Port Matilda, PA, USA). The Root Mean Squared (RMS) error statistic was used for spectral repeatability evaluation (Shenk and Westerhaus, 1995). The fat, moisture and protein composition of each sample was determined by NIRS using a robust model developed for ground Iberian meat in the range 1100-2500 nm (not published). These predictions were used as reference for further analysis.

Modified Partial Least Squared (MPLS) regression method (Shenk and Westerhaus, 1991) was used to develop calibration models for predicting fat, moisture and protein with intact pork loin samples analyzed with the fiber optic contact probe instrument. As spectral pre-treatments, Standard Normal Variate (SNV) plus Detrending (DT) (Barnes *et al.*, 1989) was used to remove the multiplicative interferences of scatter and two derivative mathematical treatments were performed: window-wise filtering (1,10,5,1) and (2,5,5,1) (ISI, 2000). The optimum number of model factors was selected by cross-validation using 4 groups.

Signal noise at the beginning and end of the spectral range was eliminated for the LabSpec®Pro spectra measurements. Two spectral regions were selected for performing the calibration models: 450-2300 nm and 1100-2300 nm.

The evaluation of the models was performed with different statistics: the standard error of calibration (SEC), the standard error of cross-validation (SECV) and the coefficient of determination for cross-validation (r^2).

III – Results

The collection of high-quality spectra is a crucial task to develop accurate calibration models. NIRS spectral repeatability using the RMS statistic can help to evaluate the quality of the spectra. In the case of the LabSpec®Pro, the RMS values exceeding the 90,000 µlog 1/R for comparisons of spectra form the same sample and location across the whole spectral range (350-2500 nm). By removing spectral areas with a very signal noise, i.e at the beginning (350-450 nm) and the end (2300-2500 nm) of the spectra, the RMS values were lowered to 86,115 µlog 1/R. Figure 1 shows the spectra collected from intact loins with the fiber-optic contact probe and it is observed the high signal noise over 2000nm. In the case of the FossNIRSystem, the RMS values are on the average of 9.941 µlog 1/R.

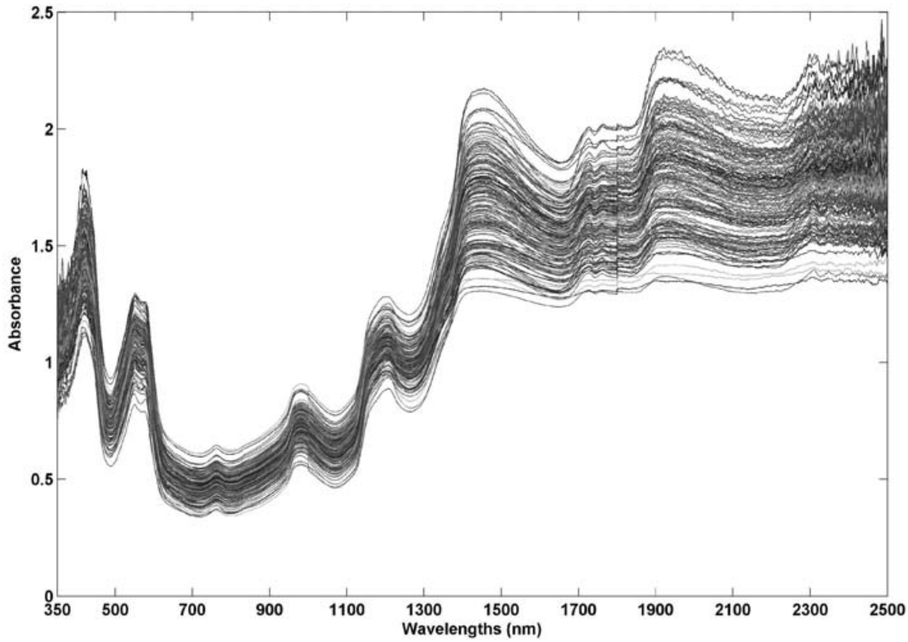

Fig. 1. Spectra collected from intact pork loins using a fiber-optic NIRS instrument from LabSpec®Pro.

Figure 2 shows the average spectra of the samples set for the analysis performed for intact pork loins with LabSpec®Pro and for ground pork meat with the FossNIRSystem instrument in the 400-2300 nm range. It is observed strong similarities between both, although the quality of the spectra is different with a smoother pattern in the case of the ground meat, due to the sample presentation and instrument differences. The FossNIRSystem is a laboratory instrument working under controlled-conditions, while the LabSpec®Pro was working on-line in the industry. Ground samples presented a more clearly-defined spectrum with sharper peaks than the intact loins. However, both show different characteristic absorption peaks: around 1200, 1720-1760nm as areas of fat and water-related peaks at 1450 and 1940 nm (Williams and Norris, 1987; Osborne, Fearn and Hindle, 1993).

The spectra of the ground samples were used to predict fat, moisture and protein composition of the Iberian pork loins with a robust model developed for the FossNIRSystem working in the

1100-2500 nm range (not published) over different years and with different ground pork muscles in our research group. Table 1 shows the statistic of that model. The prediction obtained for each sample was used as reference value for developing calibration models for the determination of fat, moisture and protein composition in intact loins using the fiber-optic instrument.

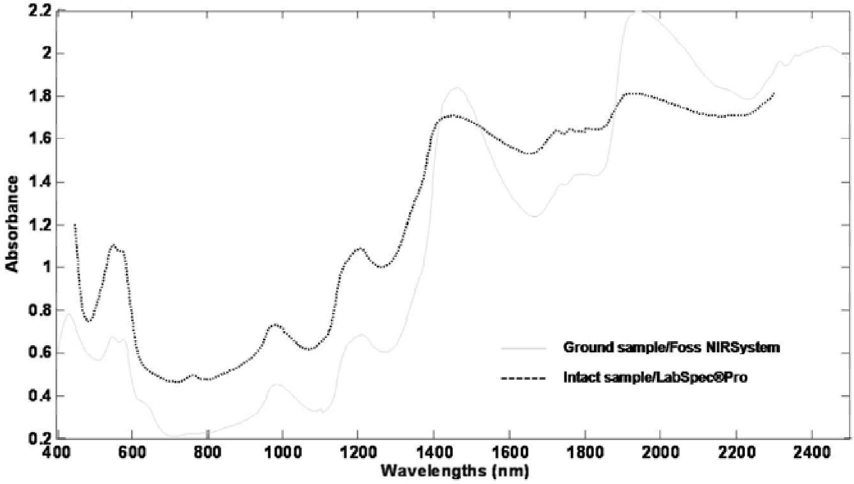

Fig. 2. Average spectra collected with the different instruments and samples presentation.

Table 1. Calibration statistics for the NIRS prediction using ground Iberian meat samples of fat, moisture and protein composition

Parameter	Pre-treatment	No. of samples	Nº of Principal Components	SEC (%)	r^2	SECV (%)
Fat	SNV + DT (2,5,5,1)	315	4	0.37	0.98	0.40
Moisture	SNV + DT (2,5,5,1)	315	5	0.47	0.96	0.51
Protein	SNV + DT (1,10,5,1)	324	7	0.48	0.91	0.49

Table 2 shows the statistics for the different parameters (fat, moisture and protein) studied of the samples set predicted by NIRS using the Table 1 models. Modified Partial Least Squared (MPLS) algorithm was used to develop calibration models for the prediction of the major chemical composition parameter in intact Iberian pork loins with a remote NIR reflectance fiber-optic contact probe instrument.

Table 2. Calibration statistics for the NIRS prediction using ground Iberian meat samples of fat, moisture and protein composition

	Training set (173 samples)			
Parameter	Minimum	Maximum	Average	Standard Deviation
Fat (%)	3.20	18.95	8.69	2.90
Moisture (%)	63.20	74.97	70.29	2.08
Protein (%)	17.16	21.95	19.68	0.90

The best calibration models obtained for predicting the three main chemical parameter in meat, measured with the fiber-optic contact probe in intact Iberian pork loin, are shown in Table 3 (i.e. after testing second and first derivative for two spectral range: 450-2300 nm and 1100-2300 nm).

Table 3. Calibration statistics of fat, moisture and protein composition obtained for the NIRS prediction with a remote fiber-optic probe using intact Iberian loin samples

Parameter	Range (nm)	Pre-treatment	No. of samples	No.º of Principal Components	SEC (%)	r^2	SECV (%)
Fat	450-2300	SNV + DT (1,10,5,1)	173	4	1.42	0.68	1.65
Moisture	450-2300	SNV + DT (2,5,5,1)	168	4	0.83	0.59	1.25
Protein	450-2300	SNV + DT (1,10,5,1)	171	4	0.51	0.56	0.59

As expected, models developed using intact loins are less accurate (Table 3) than those obtained with ground samples (Table 2). Intact loins showed larger sample heterogeneity than ground samples; freezing/drawing of the ground samples can affect mainly the moisture parameter and the quality of the spectra is different between instruments and sample presentation. Nevertheless, the models showed the possible potential of a fiber-optic contact probe NIRS instrument for analyzing chemical composition of intact pork loins. Moreover, an on-line analysis enables instantaneous and without sample presentation measurement providing important industrial advantages for quality and process control. However, there are several topics that require further studies such as the sampling optimization of intact meat products or the choice of the best instrument measurement parameters for an adequate spectra collection.

IV – Conclusions

The on-line NIRS instrument evaluated in this study for the quantitative chemical composition prediction of intact Iberian pig loins shows a very promising result enabling several practical advantages of the analysis. Further work is required in order to develop more accurate models.

Acknowledgements

The authors gratefully acknowledge the financial support provided by the research project AECERIBER “Determinaciones analíticas de muestras de carnes y grasas de razas porcinas autóctonas” and to the company Bonsai Advanced Technologies S.L. for providing the spectrometer. EZR acknowledges financial support from the Spanish Ministry of Education as a fellow of the Program “Training of University Teachers” (Formación del Profesorado Universitario, FPU).

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[1]H-NMR as a tool to determine the type of feeding of Iberian pigs

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Abstract. The fact that animals are often given feed in an attempt to achieve similar carcass characteristics to those of animals exclusively fed with "montanera", has obliged us to develop methods to differentiate and determine the type of feed consumed by these animals. One of these methods is Nuclear Magnetic Resonance (^{1}H-NMR). Iberian pig fat samples were taken in the rump at 10 cm from the tail insertion of Torbiscal pure Iberian pigs fattened with montanera (batch B1; n=44) and with feedcompound (batch P1; n=16). Similar samples were also taken from diverse genetic types of pure Iberian pigs fattened with montanera (batch B2; n=10) or with special feedcompound (batch P2; n=10). The ^{1}H-NMR spectra of these fat samples were obtained according to routine methods at the NMR Unit of the Central Service for Reseach Support (SCAI) of the University of Cordoba. The Principal Component Analysis (PCA) of ^{1}H-NMR spectral data show the possibility to determine the type of feeding of Iberian pigs by using this information.

Keywords. Iberian pig – Fat – PCA – ^{1}H-NMR.

^{1}H-RMN comme outil pour déterminer le type d'alimentation des porcs Ibériques

Résumé. *Nourrir les animaux avec des aliments composés pour obtenir des carcasses dont les caractéristiques soient semblables à celles des animaux recevant exclusivement une alimentation de type «montanera», nécessite de rechercher des techniques qui permettent de différencier et de reconnaître le type de nourriture qu'ont reçue les animaux. La Résonance Magnétique Nucléaire (^{1}H-RMN) est une de ces techniques. Des échantillons de graisse ont été prélevés dans la croupe (à 10 cm de l'insertion de la queue) de porcs Torbiscal purs engraissés selon le mode «montanera» (lot P1, n = 44) ou recevant un aliment composé (lot B1,n = 16). Des échantillons similaires ont été prélevés sur des porcs Ibériques purs de différents types génétiques engraissés selon le mode «montanera» (lot B2, n =10) ou recevant des aliments spéciaux (lot P2, n =10). Des spectres ^{1}H-RMN de ces échantillons de graisse ont été obtenus selon les protocoles d'analyse de l'Unité de RMN du Service Central d'Appui à la Recherche (SCAI), de l'Université de Cordoue. L'analyse en composantes principales (ACP) des données ^{1}H-RMN montre la possibilité de déterminer le régime alimentaire des porcs de race Ibérique à partir uniquement de ces informations spectroscopiques.*

Mots-clés. *Porc Ibérique – Graisse – PCA – ^{1}H-NMR.*

I – Introduction

Pigs used for the production Iberian pork have traditionally eaten the natural resources that abound in the Spanish countryside: fodder, grains and mainly acorns, which have fallen from the *Quercus* trees (oak, cork oak, gall-oak, etc). This system of production (named *montanera*) means that animals fattened in this way produce high quality meat and therefore meat products. The limitation of acorn production on the one hand and the high product price on the other, have led to a search for alternative products to acorns, attempting recreate the characteristics of the animals fattened on acorns and fodder, although the quality of the meat is not as good as that of animals fattened on *montanera*. In order to distinguish which carcasses belong to animals that have not been fattened on *montanera*, several analytical techniques have been developed to determine certain characteristic parameters for each feeding regime. Some of these parameters are very easy to determine, but have a high error rate, such as tactile sensation, slip temperature and iodine levels. Others, such as the determination of fatty acids, triglycerides or phospholipids, have led to a more accurate identification of the animals' feeding regime (De Pedro, 2001). However, the development of special commercial feeds that create lipids profiles in the animals similar to those provided by acorns, have cast doubt on the reliability of these techniques. Proton Nuclear Magnetic Resonance (^{1}H-NMR) spectroscopy is one of the more powerful spectroscopic tools for the investigation of the chemistry and physical properties in samples. The use of ^{1}H-NMR is becoming universal for a wide range of fields including biochemical, agricultural, medical, materials, chemical, industrial, environmental, and pharmaceutical (Alan and Alan, 2005; Larsen et *al*., 2006; Hong-Seok et *al*., 2009; Alonso-Salces et *al*., 2010). One of the benefits of ^{1}H-NMR spectroscopy is the ability to probe complex systems without necessarily requiring a separation of individual components prior to analysis. With the continued development of ^{1}H-NMR spectroscopy as an analysis tool, the size and complexity of NMR data sets make them more difficult to analyze simply through operator interaction (Alan and Alan, 2005). Multivariate methods like Principal Components Analysis (PCA) are routinely utilized in other forms of spectroscopy for the analysis of complex mixtures. The use of chemometrics in NMR is more limited, but has quickly become an important tool for the NMR spectroscopist (Alan and Alan, 2005). Hence the aim of this study is to assess how the feeding regime given to Iberian pigs affects their ^{1}H-NMR spectra and hence the possibility of using this technique to identify the animals' feeding regime, and therefore, the quality of their carcasses and their products.

II – Materials and methods

For this study, four batches of Iberian pigs were used. Two batches were fattened at the Dehesón del Encinar Research Centre, which belongs to the Department of Agriculture of Castilla La Mancha; one of them (B1; n=43 pigs) was fed exclusively on pastureland and acorns (the production of acorns was somewhat scarce), whereas the other batch (P1; n=15 pigs) was fed on commercial feed; in both cases, the fattening period was 115 days. The other two batches of animals were part of a genetic study carried out by AECERIBER in two areas of pastureland in Badajoz. One of them (B2; n=9 pigs) was fattened exclusively on *montanera* for 104 days, in which the production of acorns was abundant; the other batch (P2; n=10 pigs) was only fed commercial feed. This feed was special since one of the raw materials used was high-oleic sunflower flour, in order to product high levels of this fatty acid in the animals' subcutaneous fat. Once the animals were slaughtered, a sample of subcutaneous fat was taken from the animals' hindquarters. The sample contained skin, fat between the skin and the lean meat and a little lean meat. A liquid fat sample were extracted from each subcutaneous fat sample by using a microwave oven following the methodology explained by De Pedro *et al*., (1996). The ^{1}H-NMR spectra of liquid fat samples were obtained according to routine methods at the NMR Unit of the Central Service for Reseach Support (SCAI) of the University of Cordoba.

Each liquid fat sample was dissolved in 1 ml of deuterated chloroform and placed in a 5 mm NMR tube. The ^{1}H-NMR experiments were performed on a Bruker (Rheinstetten, Germany) Avance 400 WB spectrometer. The spectra were recorded using a 6.5 µs pulse, an acquisition time of 4.0 s (24k data points) and a total recycling time of 3.0 s, a spectral width of 3000 Hz (7.6 ppm), 16 scans. Prior to Fourier transformation, the free induction decays (FIDs) were zero-filled to 32k and a 0.3 Hz line-broadening factor was applied. The chemical shifts are expressed in d scale (ppm), referenced to the residual signal of chloroform (7.24 ppm). XWINNMR were used to perform the processing of the spectra.

The normalized spectral data were analyzed by multivariate technique like PCA with the statistical software package Unscrambler 9.2 (Camo Process AS, Oslo, Norway, 2005).

III – Results

^{1}H-NMR spectra of the 80 liquid Iberian pig fat samples were recorded. Figure 1 shows the full ^{1}H-NMR spectral range used in this work (0.5-6.0 ppm). According to this figure, a wide range of highly specific information is obtained by ^{1}H-NMR spectroscopy. The spectra include the chemical shifts of the ^{1}H signals of the different functional groups of major (triglycerides) and minor components of Iberian pig fat. Figure 2 show the score plot of the sample scores in the space defined by the two first principal components (PC1 and PC2). It can be seen that samples were grouped according to their feeding regime but some clusters were partially overlapped. It occurs with batches fed with similar type of feeding: with acorn (B1 and B2) or with commercial feedcompound (P1 and P2). However, there are more differences between batches fed with different type of feeding (B1 or B2 versus P1 or P2). To confirm these results it is suggested to test a supervised algorithm like SIMCA or Discriminant Analysis to classify these spectra from animals with different feeding. Therefore, it can be concluded that ^{1}H-NMR spectroscopy could be a useful technique to verify the animals´ feeding regime during the final fattening stage.

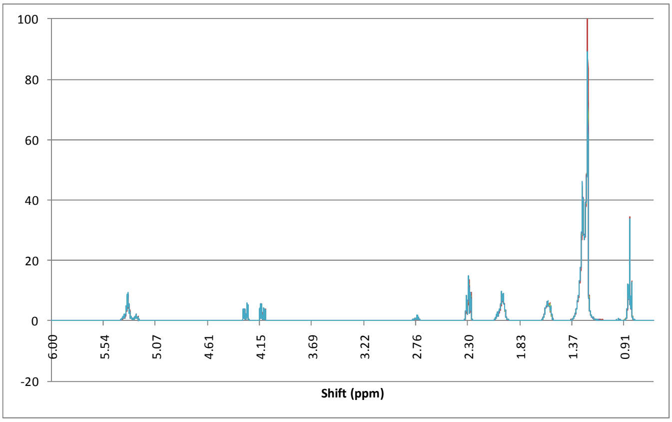

Fig. 1. ^{1}H NMR spectra of Iberian pig fat.

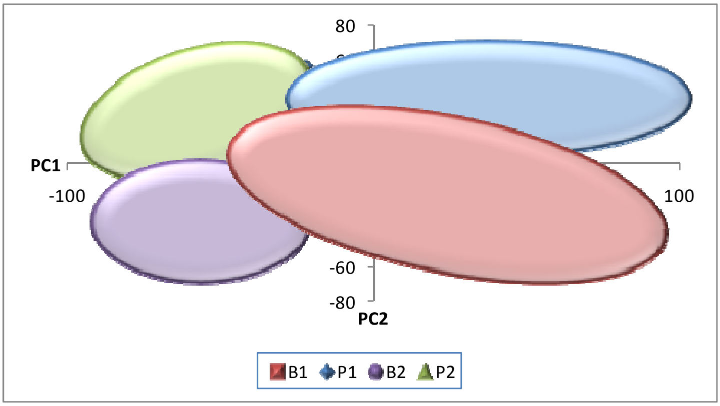

Fig. 2. Score plot.

Acknowledgements

This study has been funded by the CICYT-FEDER 1FD97-1252-C02-02 Project "Optimisation of the genetic assessment of Iberian pigs with the inclusion of quality parameters in the raw materials and in the manufactured products". The authors would like to thank the following people and organisations for their collaboration: L. Silió, C. Rodríguez, A. Fernandez and J. Rodrigáñez from the Department of Genetic Improvement of the INIA in Madrid; E. Diéguez, P. Ureta, F. Alvarez and P. Cañuelo from AECERIBER; the facilities at the "Dehesón del Encinar" Research and Experimentation Centre, which belongs to the Department of Agriculture of Castilla la Mancha's Regional Government; and the company Sánchez Romero Carvajal Jabugo, S.A.

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Acidic composition of fresh and dry-cured lard in pigs of Apulo-Calabrese (Calabrese) and Casertana ancient autochthonous genetic types (AAGT)

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Abstract. In the last fifteen years the interest in conjugated linoleic acid isomers (CLA) has increased in relation to their effect on the 'nutritional', 'extranutritional' and 'healthy' for humans. This contribute aims to use the subcutaneous adipose tissue ('lard'), 'fresh' and 'dry-cured' as a means of discriminating racial origin of 'lard'. For this aim, 20 subjects of pig ancient autochthonous genetic types (AAGT) [10 of 'Casertana' (CT) and 10 of 'Apulo Calabrese' ('Calabrese') (CL)], reared at the experimental Farm of ConSDABI Sub-NFP.I – FAO and fed a specially formulated feed not supplemented with linoleic acid (LA) and CLA, were used. The quantitative profile showed an interesting content of CLA, a significant variability depending on the genetic type and product group. CL showed a significantly higher content of CLA and linoleic acid on both 'fresh' and 'dry cured'. In conclusion it is considered that the lard ('fresh' and 'dry-cured') has to be re-evaluated to improve human health.

Keywords. Casertana – Calabrese – CLA – Ancient autochthonous genetic type – Pig.

Composition en acides gras du lard, frais et séché, des types génétiques autochtones Apulo Calabrese et Casertana

Résumé. *Au cours des quinze dernières années, l'intérêt concernant les isomères d'acide linoléique (CLA) a augmenté en lien avec leur effet sur les propriétés 'nutritionnelles', 'extranutritionnelles' et 'santé' pour l'homme. Cette contribution vise à utiliser le gras sous-cutané ('lard') 'frais' et 'séché' pour discriminer l'origine raciale du 'lard'. À cette fin, on a utilisé 20 sujets, 10 Calabresa et 10 Casertana élevés chez le ConSDABI et engraissés avec un aliment spécialement formulé non supplémenté en acide linoléique (LA) et CLA. Le profil quantitatif a montré une teneur intéressante en CLA, variable de façon significative en fonction du type génétique et du groupe de produits, qui a trait à l'étude de la technologie commerciale dans le type génétique. Le CL met en évidence une teneur significativement plus élevée en CLA et en acide linoléique soit sur le 'frais' soit sur le 'séché'. En conclusion on doit fortement réévaluer le lard ('frais' et 'séché') pour la santé de l'homme.*

Mots-clés: *Acides gras – Casertana – Calabrese – CLA – Type génétique autochtone ancien – Porc.*

I – Introduction

Linoleic acid in its conjugated form, so called CLA, was detected for the first time in the lipidic component of milk (Pariza *et al*., 2001) giving rise to a considerable interest for its documented biological effects (Matassino *et al*., 2006; Secchiari, 2006). The biological effects, as demonstrated in a wide range of animal models, are manifested in the following activities: antiatherogenic, antioxidant, anticancer, antibacterial, antiadipogenic (Lin *et al*., 1995); protection against diabetes (Belury *et al*., 2003); promoter of growth factors and immunomodulator (Hwang, 2000 ; Bassaganya-Riera *et al*., 2001, Corino *et al*., 2003). In addition to these biological effects, CLA have the ability to limit the oxidation of polyunsaturated fatty acids (PUFA) in food, reducing the phenomenon of rancidity and thus positively influencing their shelf life (Corino *et al*., 2003).

As it is known, the lipidic profile and, in particular, the content of CLA, are strongly influenced by feed regimen of the animal; even in pigs the influence of feed regimen on the qualitative-quantitative composition of the lipidic fraction was demonstrated (Dugan *et al*., 1997).

With this contribute we want to investigate fatty acid composition and the content of CLA in subcutaneous adipose tissue ('lard') in 'Apulo Calabrese' ('Calabrese') (CL) and 'Casertana' (CT) ancient autochthonous genetic types (AAGT) with the intent to detect differential biomarkers.

II – Materials and methods

The acidic fraction was extracted by the method of Folch from 40 samples of, 'fresh' and 'dry-cured' 'lard' obtained from 20 pigs (10 CT and 10 CL) raised in the same farming conditions at the experimental Farm of ConSDABI SUB NFP.I – FAO. The subsequent transesterification of fatty acids linked to glycerol was obtained by methylation of themselves with methanol and, as catalyst, potassium hydroxide (KOH). The methyl esters of fatty acids were then separated by gas chromatography with FID (flame ionization detector) and on-column injector, equipped with CP-Sil 88 column (100 m, 0.25 mm) to better separation. Fatty acid content was expressed as a percentage of total fatty acids revealed. For the assessment of the 'nutritional' value of 'lard', atherogenic (AI) and thrombogenic (TI) indexes (Ulbricht and Southgaten, 1991) were also calculated.

III – Results

1. Acidic profile

The comparison in saturated (SFA) and unsaturated (UFA) fatty acid of 'fresh' and 'dry-cured' 'lard' from two AAGT's CL and CT is reported in Table 1.

Table 1. Fresh and dry-cured lard: percentage average content of UFA and SFA and significance of the difference between the two AAGTs

Fatty acids	Fresh lard				Dry-cured lard			
	TGAA		Δ(CL-CT)		TGAA		Δ(CL-CT)	
	CL	CT	Value %	Significance	CL	CT	Value %	Significance
SFA	26.38	34.89	-8.5	0.002	29.30	35.56	-6.26	0.092
UFA	73.62	65.11	8.5	0.003	70.68	64.42	6.26	0.092

As shown in Table 1, the 'lard' of CL evidenced, regardless of the product merceological class ('fresh' or 'dry-cured'), a higher average percentage content of unsaturated fatty acids (UFA) and consequently a lower content in saturated fatty acids (SFA). The superiority of UFA in CL, compared to the CT, is highly significant for the 'fresh' 'lard' (P = 0.003) and significant near to the critical limit for the 'dry-cured' 'lard'.

The apparent decrease of UFA in the 'dry-cured' 'lard', may be related to the enzymatic mechanism of lipases, that, in swine, during ripening, act on triglycerides giving priority to positions 3 and 1, that are usually occupied by UFA, causing a decrease of the latter in their esterified form.

In both AAGTs, UFA, which have greater impact on the total UFA content are oleic and linoleic acids. As shown in Table 2, only linoleic acid was significantly different between the two AAGTs both in the 'fresh' (P <0.0001) and 'dry-cured' (P = 0.001).

The difference between the two AAGTs, was significant only for the following fatty acids: (i) in the 'fresh' 'lard': myristic acid (P <0.0001), heptadecenoic acid (P = 0.002) and stearic acid (P = 0.008); (ii) in the 'dry-cured' 'lard': myristic acid (P = 0.007) and heptadecenoic acid (P = 0.02).

The average CLA content (Table 2) was significantly higher in CL compared to CT in both 'fresh' (P = 0.002) and 'dry-cured' 'lard' (P = 0.055)

Table 2. Fresh and dry-cured lard: percentage average content of fatty acids and significance of the difference between the two AAGTs

Fatty acids	Fresh lard				Dry-cured lard			
	AAGT		Δ(CL-CT)		AAGT		Δ(CL-CT)	
	CL	CT	%	Signif.	CL	CT	%	Signif.
Lauric	0.038	0.78	-0.040	0.538	0.072	0.087	-0.015	0.971
Myristic	0.876	1.420	-0.543	<0.0001	0.978	1.464	-0.486	0.007
Palmitic	18.014	22.654	-4.640	0.812	19.408	22.714	-3.306	0.066
Palmitolenic	2.179	2.056	0.123	0.499	1.916	1.990	-0.074	0.926
Heptadecanoic	0.211	0.233	-0.022	0.693	0.225	0.238	-0.013	0.956
Heptadecenoic	0.207	0.220	-0.013	0.002	0.216	0.220	-0.004	0.020
Stearic	7.242	10.509	-3.267	0.008	8.629	10.441	-1.812	0.207
Oleic	41.229	40.346	0.883	0.667	40.115	39.318	0.797	0.987
Cis-12-C-18:1	2.254	1.704	0.550	0.045	1.509	1.525	-0.016	0.809
Linoleic	22.592	16.819	5.773	<0.0001	21.987	17.824	4.162	0.001
γ - Linolenic	0.182	0.194	-0.013	0.498	0.208	0.195	0.013	0.544
Linolenic	2.467	2.093	0.373	0.016	2.346	2.218	0.128	0.779
CLA	2.509	1.673	0.836	0.002	2.371	1.773	0.598	0.055

2. Atherogenic and thrombogenic indexes

The different fatty acid composition influenced AI and TI in the two genetic types. From Table 3 it evident that CL pig, than CT, has a significantly lower value of both indices. The values measured for CL are in agreement with those reported by other authors (Matassino *et al.,* 2005) while CT values are slightly higher (Matassino *et al*., 2008).

Table 3. Fresh and dry-cured lard: atherogenic and thrombogenic indexes and significance between the two AAGTs

Indices	Fresh lard				Dry-cured lard			
	AAGT		Δ(CL-CT)		AAGT		Δ(CL-CT)	
	CL	CT	Value %	Signif.	CL	CT	Value %	Signif.
Atherogenic	0.303	0.448	-0.145	0.006	0.342	0.453	-0.110	0.093
Thrombogenic	0.726	1.074	-0.348	0.001	0.837	1.077	-0.240	0.002

IV – Conclusions

The research showed that some fatty acids can be used to discriminate, whith probabilistic approach, both 'fresh' and 'dry-cured ‘lard’ provided by a given pig AAGT.

In addition, the ‘lard’ has an atherogenic and thrombogenic index lower in comparison with other animal and plant origin products discretiting many preconceptions about its inclusion in a human food regime.

Acknowledgements

The authors wish to acknowledge the financial support received from Ministry of Agricultural, Food and Forestry *(* Mipaaf).

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Individual characterization of Iberian pig through NIRS technology: Implementation in Sierra de Sevilla, S.A.

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Abstract. The Iberian pig industry has established several quality control programs in order to determine and guarantee the pig feeding system, especially in the last period of growing. These programs consisted of on-farm Iberian pig inspection or fatty acids composition of subcutaneous tissue fat. The potential of NIRS technology in the classification of Iberian pig carcasses in the several commercial categories, according to their feeding system, has been reported by researchers at the University of Cordoba in the last years. However, the implementation of this technology at industrial level has not been yet as widespread as expected in comparison with other industries such as animal feed industry. This communication reports the results obtained in the first year of the project for the implementation of NIRS technology as quality control tool in the "Sierra de Sevilla" Company which elaborates meat products from Iberian pig. The use of NIRS technology as quality control tool allowed a fast, reliable and inexpensive characterization of each animal. Individual characterization is of great importance in "recebo" batches, where a large variability between animals was found. The good results obtained to date have given individual information of its animals to the Company providing a better quality control of their products and also monitoring the farm suppliers.

Keywords. Iberian pig – Agro food industry – NIRS technology – Quality control – Feeding system.

Caractérisation individuelle du porc Ibérique par la technologie NIRS : Mise en œuvre dans l'entreprise "Sierra de Sevilla" S.A.

Résumé. *L'industrie du porc ibérique a établi des programmes de contrôle de qualité pour déterminer et veiller sur le système d'alimentation des porcs, en particulier dans la dernière période de croissance. Ces programmes comprennent l'inspection des animaux sur le terrain et l'analyse de la composition en acides gras des tissus adipeux sous-cutanés. Des années de recherche à l'Université de Cordoba ont montré le potentiel de la technologie NIRS pour la classification des carcasses de porc Ibérique en catégories commerciales, en fonction de leur système d'alimentation. Toutefois, en comparaison avec d'autres industries (par exemple, celle des aliments pour animaux), la mise en œuvre de cette technologie au niveau industriel pour le secteur du porc Ibérique est encore très limitée. Cette communication présente les résultats obtenus dans la première année du projet pour la mise en œuvre de la technologie NIRS comme outil de contrôle qualité dans l'entreprise de produits à base de viande provenant de porcs Ibériques «Sierra de Sevilla». L'utilisation de la technologie NIRS comme outil de contrôle de qualité a permis une caractérisation rapide, fiable et peu coûteuse de chaque animal. La caractérisation individuelle est d'une importance cruciale dans les groupes du «recebo», où une grande variabilité entre les animaux appartenant au même groupe a été trouvée. Les bons résultats obtenus à ce jour ont donné des informations individuelles sur les animaux de l'entreprise, en permettant un meilleur contrôle de la qualité de leurs produits et aussi un meilleur suivi des élevages fournisseurs.*

Mots-clés. *Porc Ibérique – Industrie alimentaire – Technologie NIRS – Contrôle de la qualité – Système d'alimentation.*

I – Introduction

Iberian pig ham is one of the most expensive food products produced in Spain. Three commercial categories ("*Bellota* or Acorn", "*Recebo* or Acorn+compound feed" and "*Cebo* or compound feed") are established, according to the pig feeding system, especially during the final phase of the growing period. The classification of these expensive products is of huge interest for industries, authorities and consumers and, for this reason, great efforts have been performed to obtain efficient and reliable methods that allowed a right and unequivocal classification (De Pedro, 2001). In the last years, different methods were applied on the basis of the fatty acid profile in subcutaneous fat or on-farm inspections to classify pig batches in commercial categories (Garrido-Varo and De Pedro, 2007); however these methods provided global information of each batch but individual information of each pig is missing. On the other hand, consumers would have as much nutritional information as possible of these expensive food products and Iberian pig industry has to meet consumer's requirements in order to guarantee the quality of these products and thus satisfy the consumer requests.

The results obtained in the first year of the implementation of NIRS technology as quality control tool in "Sierra de Sevilla" Company allowed a fast, inexpensive and reliable characterization of each pig according to their fatty acid profile. As a result, "Sierra de Sevilla" could improve the maturation process of each piece as well as the quality assurance and traceability of each one of its food products.

II – Materials and methods

A set of 250 Iberian pig subcutaneous fat samples, from 17 batches with different pig feeding system, were analysed by NIRS in Sierra de Sevilla (Sevilla, Spain) and official gas chromatography data were obtained from a certified laboratory.

NIR absorbance spectra were collected in reflectance mode (log (1/R)) using a FOSS-NIRSystems 5000 spectrometer equipped with a remote reflectance probe (FOSS, Hillerød, Denmark), in the wavelength region between 1100 and 2500 nm (every 2 nm). Each subcutaneous fat sample was taken from carcass of Iberian pigs in the sacrifice line, from the tail insertion area in the coxal region, the same area as that used by the Designation of Origin committees (ref). Subcutaneous fat samples were stored at –20 ºC until analyses were performed. NIRS analysis was carried out by placing directly the thawed subcutaneous fat sample on the probe and spectra were collected and recorded using ISIscan™ Routine Analysis Software 3.5 version (Infrasoft International, Port Matilda, PA, USA).

Once the NIRS analysis was performed, adipose tissue sample was heated by microwave energy to melt the fat (De Pedro *et al.,* 1997). After removing the supernatant subcutaneous tissue, the appropriate liquid sample amount was taken to be sent to the laboratory, where the fatty acid composition was determined by gas chromatography, in accordance with the official method, to get the reference data.

NIR and reference data were used to develop modified partial least squares (MPLS) calibration equations by using WINISI software version 1.50 (FOSS NIRSystems Inc., Laurel, MD, USA) to predict fatty acid profile of the fat tissue samples (Williams & Sobering, 1996).

III – Results and discussion

As animal with different feeding systems had been selected, individual reference data of the 250 samples showed a great variability, (i.e. oleic acid ranged 48 to 58%), and so enough variability has been included in the calibration model to provide reliable predicted NIRS data in all categories.

Table 1 and Table 2 show individual fatty acid profile (4 major fatty acids) of 12 Iberian pigs from a batch of 130 animals fed with grass and acorns (*bellota*) and another 12 Iberian pigs from a batch of 120 animals fed with grass, acorns and supplemented with compound feed (*recebo*), respectively.

Table 1. Gas chromatography and NIRS predicted data of the 4 major fatty acids in a *bellota* batch

Sample	Palmitic acid (C16:0)		Stearic acid (C18:0)		Oleic acid (C18:1)		Linooleic acid (C18:2)	
	LAB	**NIR**	**LAB**	**NIR**	**LAB**	**NIR**	**LAB**	**NIR**
1	21.5	21.4	9.2	9.7	54.9	54.2	8.6	8.5
2	20.5	20.7	9.0	9.3	55.9	55.3	8.9	8.8
3	19.6	19.9	9.7	9.4	56.1	56.2	9.9	8.8
4	19.9	19.7	8.3	8.5	56.6	56.3	9.2	9.2
5	19.3	19.5	8.7	9.2	57.1	56.4	9.5	9.2
6	19.2	19.9	8.3	8.5	57.5	56.8	9.2	9.3
7	18.7	19.4	8.3	9.5	57.7	57.5	9.8	9.1
8	19.1	19.6	8.3	8.1	57.9	57.3	9.2	8.9
9	19.0	19.7	7.6	8.0	57.9	56.6	9.6	9.7
10	19.0	18.9	8.7	8.2	57.9	57.6	8.9	9.2
11	18.8	18.9	7.7	7.9	58.3	57.9	9.6	9.5
12	19.2	19.4	8.4	8.0	58.5	58.3	8.4	8.8
…	…	…	…	…	…	…	…	…
Lab/NIR average (n=12)	19.5	19.8	8.5	8.7	57.2	56.7	9.2	9.1
Lab/NIR Variance (n=12)	0.7	0.5	0.4	0.4	1.1	1.2	0.1	0.1
Batch average (n=130)	19.3		8.1		56.8		98	

As can be seen in both Tables, slight differences between official gas chromatography method and NIRS predicted values were found in all samples (12 Iberian pig of each batch) as well as in average values, indicating the good prediction ability of the calibration model. The batch average value, usually employed in the Iberian pig industry to classify Iberian pigs and select the best maturation process of pieces (hams, shoulders and sausages) is also indicated at the bottom of the table.

Individual *versus* batch average analysis of Iberian pig subcutaneous tissue samples allowed detecting differences in the fatty acid profile of animals from a same batch, being these differences more marked in the *recebo* batches than in *bellota* ones. So, oleic and stearic acid values from 50.3 to 57.8 and 8.3 to 11.7, respectively, were found in the *recebo* batch (see Table 2) whereas in the *bellota* batch (see Table 1) these fatty acids only varied from 54.8 to 58.5 and from 7.6 to 9.7, respectively. In general, the magnitude of these differences between animals from a same batch of *bellota* or *recebo* could be observed by using statistical parameters to compare sample sets such as coefficient of variation, standard deviation or variance, among others.

Tables 1 and 2 show variance values found for each fatty acid in *bellota* and *recebo* batches, respectively. In all cases, variance values are higher in *recebo* than in *bellota*, owing to the greatest variability between animals from *recebo* batch.

Table 2. Gas chromatography and NIRS predicted data of the 4 major fatty acids in a recebo batch.

	Palmitic acid (C16:0)		Stearic acid (C18:0)		Oleic acid (C18:1)		Linooleic acid (C18:2)	
Sample	**LAB**	**NIR**	**LAB**	**NIR**	**LAB**	**NIR**	**LAB**	**NIR**
1	22.9	22.7	12.0	11.7	50.4	50.3	9.0	8.7
2	21.5	21.8	9.9	10.5	52.9	53.8	9.8	9.1
3	21.5	21.3	10.3	10.5	53.4	53.8	8.9	8.6
4	21.5	21.2	9.9	9.7	53.6	54.8	8.7	8.7
5	21.3	21.3	10.7	10.7	53.7	53.1	8.4	8.4
6	20.0	20.1	9.4	9.0	55.0	55.2	9.7	9.3
7	19.2	20.1	10.7	10.1	55.8	55.5	8.6	8.9
8	18.8	19.6	9.6	8.9	56.5	56.1	9.3	9.2
9	19.0	19.4	8.6	8.4	57.0	57.2	9.1	9.0
10	18.9	19.5	8.3	8.2	57.1	57.2	9.4	9.4
11	17.6	17.8	9.8	9.1	57.8	57.4	9.2	9.8
12	18.8	18.3	9.5	9.2	57.2	57.3	9.3	9.6
…	…	…	…	…	…	…	…	…
Lab/NIR average (r=12)	20.1	20.3	9.9	9.7	55.0	55.1	9.1	9.1
Lab/NIR variance (r=12)	1.4	1.2	0.6	0.6	2.2	2.0	0.2	0.2
Batch average (n=120)	19.4		9.4		55.2		9.8	

Taking into account that Iberian pig industries use the 4 major fatty acid values of the batch as quality control parameter for all the animals included in this batch, the implementation of NIRS technology in Sierra de Sevilla, S.A. for the individual characterization of each animal implies a step forward in the quality assurance and traceability of their food products.

IV – Conclusions

(i) NIRS technology allows a fast, inexpensive and reliable characterization of each pig according to their fatty acid profile of the subcutaneous tissue, providing similar results as the official gas chromatography method.

(ii) Individual *versus* batch average analysis could detect differences between animals included in the same batch, being these differences more marked in the *recebo* batch.

(iii) The implementation of NIRS technology in Sierra de Sevilla S.A. enables individual quality control in their production process monitoring each animal at the reception zone and giving useful information for their optimal maturation process.

Acknowledgments

This research is a part of the Project "Implantación de la Tecnología NIRS en la caracterización de canales y productos derivados de cerdo Ibérico en la empresa Sierra de Sevilla, S.A.", funded by the "Consejería de Innovación, Ciencia y Empresa" of the Junta of Andalucía.

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Session 7
Socio-economic aspects

Parma PDO Ham in pork production chain and in Parma economy[1]

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Abstract. Prosciutto di Parma DOP (Parma PDO Ham) is the main product of Italian cold cuts and is produced in the province of Parma. The verification of compliance with its guidelines is implemented by Istituto Parma Quality (IPQ), that together with Istituto Nord Est Qualità (INEQ) operates a monitoring on compliance requirements of the most part of Italian PDO and PGI cold cuts production. In the production area there are 181 production plants where 9 milions of PDO Ham are produced every year and the 71% of factory and the 80% of Parma PDO Ham production is concentrated in four municipal districts in the foothills of the province of Parma. Among the major economic and financial aspects it is to consider production difficulties related to the duration of financial cycle because long maturing period and because of long terms of payment required by large retail chains. Analyzing a sample of firms it emerges the improving technology level and the management strategies with the aim to differentiate production (Parma Grosso ham production), and pre-sliced ham production. These strategies make it possible to generate positive cash flows to serve indebtedness and to reward equityholders.

Keywords. Parma PDO Ham – Pork production chain – Cash flow analysis – Parma economy.

Le Jambon de Parme AOP dans la chaîne de production porcine et dans l'économie de Parme

Résumé. *Le Prosciutto di Parma DOP (Jambon de Parme AOP), produit dans la province de Parme, est le produit le plus important de la charcuterie italienne. Le contrôle du respect du cahier des charges est assuré par l'Istituto Parma Qualità (IPQ), qui, avec l'Istituto Nord-Est Qualità (INEQ), surveille la conformité de l'origine d'une grande partie des AOP et IGP de la charcuterie italienne. Dans le domaine de la production il y a 181 installations qui produisent 9 millions de jambons AOP par an; sur le territoire des quatre municipalités de la province de Parme au pied de la colline sont situés 70% des établissements de fabrication et 80% de la production de Prosciutto di Parma DOP. Parmi les principales difficultés économiques et financières du secteur se trouve la durée du cycle de financement dû à la longue période de maturation et aux longs termes de paiement de la distribution. L'analyse de l'échantillon d'entreprises enquêtées montre la stratégie visant à améliorer la technologie et le processus de gestion, et aussi la variation de la production (production de jambon Parma Grosso), et la production de jambon pré-tranché. Ces stratégies permettent de générer des flux de trésorerie positifs pour payer la dette et de récompenser les détenteurs du capital-risque.*

Mots-clés. *Jambon de Parme AOP – Chaîne de production du porc – Flux de trésorerie – Économie de Parme.*

I – Introduction

In the province of Parma there is an important production chain related to processing of pork meat to produce Prosciutto di Parma DOP (Parma PDO Ham): in the province could be considered others cold cuts productions as Culatello di Zibello DOP, Salame di Felino, Spalla Cotta di San Secondo Parmense, Coppa di Parma, and other productions as not marked hams, with less weight and lower period of maturing. In general, in Italy the pig sector is characterized

[1]The work, although the result of a joint reflection, was prepared as follows: Giuseppe Bonazzi wrote paragraphs 1, 2, 4, 7, Mattia Iotti wrote paragraphs 3, 5, 6.

by the herd of swine heavy pig (weight of 160 ± 10 kg) as particular animal that is bred to produce pigs to be processed for typical Italian cold cuts products, especially the typical ham (PDO ham); for these hams are used fresh legs of pigs born, raised and slaughtered in a defined area; these pigs must have quality characteristics defined by specific production rules. According to ISTAT data, the Italian production of pigs in 2009 consists of 12,922,000 animals, of which 8,707,362 are to produce PDO (IPQ-INEQ data) cold cuts. The slaughtered animals are 13,593,774 of which 671,774 came from foreign countries. The consumption of pork, in Italy, is 37.68 kg per capita in 2009 so the rate of self-supply in Italy was 68.9%. Genetic selection operates with two separate addresses: for the cold cuts production, it is used Italian Large White (LWI), Italian Landrance (LI) and Italian Duroc (DI) even for production of meat for butcher is used race Petrain (P). It was conducted an important activity for the conservation of native pig breeds, to protecte genetic types as Cinta Senese, Mora Romagnola, Nero siciliano, Casertana, Apulo-Calabrese and Sarda. This selection has given the availability of animals with proper genetic definition and with clear breed characteristics for commercial enhancement. The breeding program for the heavy pig is based on the rearing of offspring of boars Italian Duroc and Large White sows for Italian Landrance. The selection objectives are to achieve an efficient conversion of feed, lower losses in livestock, no defects at slaughterhouse to increase production efficiency, plain carcass composition in order to improve meat quality for producing PDO hams and matured meats. The consumption of cured meats in Italy in 2009 was 1.1745 million tons; the ham is the first cold cuts for consumption in Italy, with 280.6 thousand tons, then we have cooked ham, with 275.8 thousand tonnes, Mortadella, with 173.9 thousand tons, and Salame, with 110,4 thousand tonnes. In the Italian cold cuts has an important role in the protected products: in Italy production of cold cuts recognized with PDO and PGI are 33, with a higher concentration in northern regions. In 2009 there were 8,680,611 slaughtered pigs certificates PDO, 17,361,222 available thigh for PDO production, 14,550,654 thighs started to PDO production, of which 9,429,462 for the PDO Prosciutto di Parma and 2,521,213 for the PDO Prosciutto San Daniele.

II – The PDO chain

The European Community, considering that production, processing and distribution of agricultural products and foodstuffs play an important role in the Community, has implemented a strategy of diversification of farm production in order to achieve a better balance between supply and demand in the markets. Therefore, Reg. (EC) No 510/2006 of 20 March 2006 regulates the protection of geographical indications and designations of origin for agricultural products and foodstuffs. The verification of compliance with specifications (Article 11) shall be performed before selling the product on the market by one or more competent agency and / or one or more control agency within the meaning of art. 2 of Reg (EC) No 882/2004 that operates as a product certification institution. The certification agency of products act in compliance with European standard EN 45011 or ISO / IEC Guide 65. The Istituto Parma Qualità (IPQ), joined with the Istituto Nord Est Qualità (INEQ) has implemented a system that provides control and compliance requirements for origin of raw materials and production process upstream in the chain. In this system of rules and controls, the farms must put the firm code and the month of birth code on both legs, in order to have the slaughter of animals with at least nine months of life; in this way it is possible within thirty days after the birth of the pig to exclude animals born outside the territory of origin. The transfer of animals between farms should be documented so it could be easier the control issued by IPQ and INEQ, even having the pigs supply control. In 2008, the herds for PDO production were 4,819 in 11 regions of central and northern Italy. The highest concentration of farms is 1,936 farms in Lombardia, then we have Piemonte (970) and Emilia Romagna (926), so that 79.5% of herds is located in these three regions. For the distribution of pigs per genotype, on 2008 data, there is the prevalence of pigs from hybrid verro, representing 67.2% of the total, followed by of mixed blood of pigs boars of other breeds with 19.1% value.

The card is also sent to IPQ-INEQ that monitor and insert the data in the respective database. The slaughterhouse must fill out a document for each day of production with a list of all lots of animals received and the number of pigs slaughtered, codes of origin and origin. Moreover, the slaughterhouse puts on each thigh a stamp of approval attesting to the code compliance of origin, provenance and quality.

Slaughterhouses active in this system in Italy (year 2008) are 121 and have slaughtered more than 9 million pigs, the largest number of slaughterhouses has in Lombardia (38), Emilia Romagna (27) and Piemonte (18). All slaughterhouses are inspected at least once a year to control information in the slaughterhouses document that must be prepared every day and of which a sample is detected as for the incoming streams of live pigs and outflows of raw material. This procedure, associated to a selection, allows the exclusion of the fresh pork legs not suitable for production and to keep down the final index of non-compliance. It is to note that slaughtered pigs must be accompanied by a document indicating the code of the firm of origin, the number of pigs, the genetic type and destination.

The fresh raw material arrives at the cold cuts production firm with the stamp of identification and self-certification of slaughter, with a copy of the document issued by the slaughterhouse. The production firm, for each delivery, analyzes the conformity of raw material and mark in an official register all description and identification elements of the product. The raw material is then initiated for processing considering that during the whole process production firm has to ensure that "ID cards and descriptive production package"; IPQ and INEQ include these data in their database and check all the documents produced for each lot of production. It is the responsibility of IPQ and INEQ to check all the quality standards of the cured product by testing and verifying the minimum maturing period, the absence of morphological, technical and taste defects. After these procedures, PDO certification mark is applied to product and it could be marked on the product label.

III – Parma PDO Ham

The Parma Ham is the most important production of Italian cold cuts sector. In 1963, 63 producers founded Parma Ham Consortium (Consorzio del Prosciutto di Parma) that today represents 164 processing firm. The Consorzio del Prosciutto di Parma manages and protects production rules, is responsible for economic policy management in the sector, supervises and ensures the respect of laws and regulations such as the protection of the name "Prosciutto di Parma" and its brand; Consorzio del Prosciutto di Parma is also the guide of trade policies and has the role to enhance the product, conducting advertising actions and fairs to assist firms.

Parma PDO Ham according to EEC Regulation 2081/92 (now Regulation EC 510/06) is produced observing production regulations issued by the Consorzio del Prosciutto di Parma, so Parma PDO Ham is obtained by processing thighs of heavy pig that must be older than 9 months of age, weighing over 150 kg that must be slaughtered "healthy, rested and fasted for at least 15 hours", as per the specification rules.

The pig must be reared in the territory of 10 regions of northern and central Italy but the production process must be done in one part of the province of Parma between the Via Emilia, at a distance of at least 5 km from here, from north, by the river Enza, to the east, and by the river Stirone, from the west. Towards the south we have a limit of production that is an altitude above sea level exceeding 900 meters. in addition to the Consorzio del Prosciutto di Parma, Istituto Parma Qualità (IPQ) conducts necessary checks on the ham as "third and independent part". IPQ performs the functions of control over the ham from 1998 and the activity is performed through unified services control together with Istituto Nord Est Qualità (INEQ) that is this agency involved in Prosciutto di San Daniele PDO control, under a system of division of control activities in the sector of the PDO.

As regards the breakdown by size classes of manufacturing, based on the hams established to production of Parma PDO Ham, there is a concentration of production in a small number of firms (Table 1); with a total production analyzed of 9,429,642 hams processed in 181 plants, with an average production of 52,096 hams per plant, 58.85% of production is concentrated in 25.41% manufacturing plants that are characterized for an annual production of more than 100,000 hams. Manufacturing plants with less than 25,000 hams per year produced 5.62% of the hams, involving 34.25% of plants; moreover it is to note that the plants up to 1,000 pieces per year of production are 8.84% of the total (0.08% of production), while the plants up to 10,000 pieces per year of production are the 22.10% of the total, with only 1,14% of the number of Parma Ham in 2009.

Table 1. Production plant per rank size (2009)

Production plant per rank size	Hams (no.)	Hams (%)	Production plants (no.)	Production plants (%)
0 – 1,000	7,652	0.08	16	8.84
1,001 – 10,000	136,870	1.45	24	13.26
10,001 – 25,000	384,970	4.08	22	12.15
25,001 – 50,000	2,067,900	21.93	53	29.28
50,001 – 75,000	1,282,433	13.60	20	11.05
75,001 – 100,000	2,161,614	22.92	25	13.81
100,001 – 200,000	2,316,990	24.57	17	9.39
> 200,000	1,071,033	11.36	4	2.21
Total	9,429,462	100.00%	181	100.00

Source: IPQ.

The consumption of Parma PDO Ham is done for 79% on domestic market and 21% in foreign markets; 2,046,495 hams are exported in 2009 (12,662 tonnes) with an estimated turnover of 181 million euro. In the foreign markets France and the United States prevail, but the European market accounts for 75.05% of exports, while the American continent is 21.30% of exports, of which 18.81% in the USA alone; exports to other states are modest, except Japan, that accounts for 4.28% of exports, approximately 87 thousand hams in 2008. In recent years there has been an increase in consumption of Parma ham sliced and packaged in boxes for sale in the refrigerated counter. During the period 2005/2009 the increase in the number of meats sliced was equal to 83.2%, from 627,344 to 1,149,574 and the relative packages production increased from 30.885 million of 2005 to 54.796 milion of 2009. The slicing process performed in the production chain make easier the consumption process, particularly in foreign markets where the process of slicing made at the store or directly from the consumer is not always carried out with the necessary expertise, thus penalizing the sale to final consumer. With regard to market, Parma sliced ham, with a total production of 6,010,930 kg of food (1,865,490 kg for domestic consumption, up to 31.03%, and 4,145,440 kg, 68.97%, for export) confirms the presence of foreign demand for a product with a high level of service. Even with respect to exports of sliced Parma PDO Ham, there is a concentration of demand in some foreign markets, so the top 5 export destination markets are Britain, France, Belgium, Germany and the USA; these markets generate 79.41% exports (49.12% for the first two target markets). Even if the sliced product is concentrated in European market, that consumes 86.55% of exports, is also of relevance the USA market for the sliced product (8.83% of the export market for sliced), so other target markets are, on 2008 data, of marginal importance.

IV – Prosciutto di Parma DOP in the local socio-economic system

The activities related to agriculture, processing industry and related services are important in the socio-economic system in the province of Parma. The food industry is the first industry in the province, with 2008 sales of 7,500 million euro, 36.6% of total industrial sales in the province, deriving € 973 million from exports. The turnover of the mechanical food plant industry is the third largest industrial sector of the province, behind the general mechanical industry, with a level of 2,200 million euro as per 10.7% of total industry turnover. The sectors of the food and plant for food, taken together, amounted to 47.3% of the industry turnover of the province.

Within the food sector with the highest turnover is the sector of pasta, bread, pastries, frozen foods and related products, as per including the presence of some large companies, that is, in 2008, 3,000 million euro turnover (40.0% of provincial revenues in the food industry and to 14.6% of industry turnover in general). The meat preserved industry generates 900 million euro turnover in 2008 (25.3% of sales in the food industry and 9.3% of industry sales in general). The province of Parma is characterized by a important meat processing activities so the local socio-economic system is therefore characterized by the presence of a large number of firms in processing pork meat for the production of cold cuts. As for the meat processing industry, based on data made available by the Registry of firms at the Chamber of Commerce of Parma, updated at August 2010, are operating 446 businesses and 143 local units of enterprises operating in the meat processing industry in the province of Parma (10.13 main activity code according to the classification ATECO 2007), as for a total of 589 units in the sector in the province of Parma. The firms involved in the sector give job to 4,399 staff employees and 322 independent operators, for a total of 4,721 person. The municipalities in the province with greater presence of these firms are Langhirano (123 companies, 41 local units, 1,140 staff employees and 72 independent operators), Lesignano de' Bagni (38 companies, 10 local units, 352 staff employees, 18 independent operators) Felino (35 companies, 17 local units, 634 staff employees, 44 independent operators), Sala Baganza (26 firms, 17 local units, 609 staff employees, 19 independent operators). The municipality of Parma has 38 companies, 10 local units, 366 staff employees and 21 independent operators.

As for Parma PDO Ham production for the year 2009, out of 181 factories in the municipality of Langhirano were processed 4,032,198 hams, representing 42.76% of the total, in 79 plants, equal to 43.65% of the total. In the first four areas of production (Langhirano, Lesignano de Bagni, Sala Baganza and Felino) it has been done the 80,28% of production (Table 2).

Table 2. Production plant per municipality (2009)

Municipality	Production plants (no.)	Production plants (%)	Hams (no.)	Hams (%)
Langhirano	79	43.65	4,032,198	42.76
Lesignano de' Bagni	22	12.15	1,480,612	15.70
Sala Baganza	16	8.84	1,134,104	12.03
Felino	12	6.63	922,619	9.78
Others	52	28.73	1,859,929	19.72
Total	181	100.00	9,429,462	100.00

Source: IPQ / Unione Parmense degli Industriali.

The development of the meat processing sector has encouraged the local employment and the creation of firms active in related production of meats, including craft processing services, production control activities, construction of facilities and machinery activities for meat processing, industrial activities and building, warehousing, as well as study and research activities involved in meat processing process. In the province of Parma there are the University of Parma with the Faculty of Agriculture, Degree in Food Science and Technology, Veterinary Medicine, Economics and Engineering, particularly focusing on applied research in the meat processing industry. There are also the Experimental Station for the Food processing and preservation (Stazione Sperimentale Industria Conserve Alimentari, as SSICA) as a public applied research center with the purpose of scientific progress in the areas of the Italian canning fruits, vegetables, meats and fish. In Parma there is also the the European Food Safety Authority (EFSA) headquarter, as the European Union agency aiming to control food safety and other agencies involved in supply chain control as Istituto Parma Qualità (IPQ).

V – Economic and financial firm data in the Parma PDO Ham industry

In the area of Parma, ham firms often have difficulties relating to the duration of the financial cycle, because the firms require large investments in start-up activity (Bonazzi *et al*., 2007), for the acquisition of industrial buildings, plants and equipments. This necessity is inherent with the typical production of ham, which requires large volumes for processing and maturation, and then expanding the need for capital equipment. In addition, the cycle of maturation of the pork leg causes a further expansion of capital requirements in order to sustain the cycle of working capital. Finally, given the sales channel frequently used by firms in the sector for market access, namely large-scale distribution (GDO), there is an increase in average day extension of credits (even this aspect of the financial dynamic improve capital requirement for processing firms). This situation quite often expands the duration of financial cycle in which, even in the face of positive profitability, it is to note unsustainable situations in terms of generating cash flow (unlevered free cash flow) available for debt service; it then becomes important in the sector (Bonazzi *et al*., 2007) a strategy to contain costs of production, even realizing investments in technologies that could reduce processing cost of raw materials (such as the activities of boning and greasing ham). In order to evaluate these investments, it is possible to use methods of assessment based on values deriving from accounting data (Roi, Roe, Rod), integrating these with assessment methodologies based on cash flow analysis (Npv, Irr, Pbpa), and methodologies to evaluate the business cycle as sustainable (Dscr, Adscr, Llcr). The Parma PDO compartment expresses case of differentiation strategy in production firms; In fact (Bonazzi *et al*., 2008) there are firms able to diversify its production by having attention to raw material quality also working with a large size choice of pork legs. These firms have production of Parma PDO Ham Parma of high category level, weighing over 9.0 kg (24 maturing months). In this way, firms even with higher production costs for raw materials could be able to obtain on the market a selling price higher than production costs.

VI – The sample of firms

The analysis of annual accounts (year 2008) of a sample of 40 firms in the area of Parma Ham is conducted by analyzing the balance sheet and income statement, on the basis of data made available by the local Chamber of Commerce. The analysis of profitability of business management is performed having the analysis of annual economic accounts (Ferrero, *et al*., 2005). The annual accounts analysis considers the data presented in the table required by law (Andrei *et al*., 2006), in patterns defined by the European Union, in accordance with the Fourth EU Directive, in order to make the different firms data comparable even if deriving from different European countries (Andrei *et al*., 2006). It is therefore useful to analyze the annual accounts data according to reclassification exposure schemes (Iotti, 2009) that aggregate data to increase provided information level, marking additional capital and profit margins (Ceccacci *et al*., 2008). The reclassification of the income statement using the value-added scheme (Bonazzi *et al*., 2005) is:

$$S \pm \Delta I - C_e = VA - C_w = EBITDA - (D + A) = EBIT \pm I \pm V \pm E \pm T = \Pi \quad (1)$$

In (1) S is sales, I is the stock (inventory), C_e is external costs, VA is value added, C_w is the cost of labour, EBITDA is earnings before interest, taxes, amortization, and depreciation, D is depreciation, A is amortization, EBIT is earnings before interest and taxes, I is interest, V is revaluations and depreciation, E is extraordinary income or expenses, T is income tax, Π is profit. The reclassification of the balance sheet (Ceccacci *et al*., 2008) is conducted according to liquidity level:

$$TA = \alpha WC_t + FA = \alpha WC_c + \alpha WC_i + \alpha WC_{ar} + FA \quad (2)$$

In (2) TA is total asset, αWC_t is total investment in working capital, αWC_i is working capital in inventories, αWC_C is working capital in cash, αWC_{ar} is working capital in account receivables, FA is fixed assets. Reclassification of balance sheet liabilities is conducted according to the origin of sources of capital:

$$TS = E + D = E + \beta WC_t + DF_s + DF_l \quad (3)$$

In (3) TS total liabilities and shareholders equity, E is equity, D is total debt, βWC_t is total working capital liabilities, DF_s is short-term liabilities, borrowings (due within 12 months), DF_l is a medium/long term financial debt (duration 12 months). The difference between αWC_t and βWC_t is net working capital (NWC). The firm income considers the accrual basis (Andrei *et al*., 2006) and expresses the moment of creation of value so the income statement is not dependent by the generation of cash flow from operations. It is therefore useful to consider the annual account because this document could be useful to analyze different sources of cash flows (Shireves *et al*., 2000). To quantify the cash flows could be used an indirect approach, through the cash flows statement (Brealey *et al*., 2003) in order to calculate unlevered free cash flow and free cash flow to equity:

$$EBIT + D + A \mp T = CF + \Delta^{\mp} NWC = OCF + \Delta^{\mp} FA = UFCF - DS = FCFE \quad (4)$$

In (4) CF is cash flow, OCF is operating cash flow, UFCF is unlevered free cash flow, FCFE is the cash flow available for shareholders (free cash flow). CF is EBIT corrected with costs that do not cause an outflow of money (D + A) and the impact of income taxes (T), OCF quantifies the absorption of net working capital (NWC) and has, in the case of Parma ham firms, a particular importance for the impact of investments in maturing inventories (hams); UFCF is determined as the sum of OCF and the absorption of capital resulting from investments in fixed assets (FA), as to say that UFCF is the cash flow available for debt service (DS, debt service, defined as DS = K + I, where K is the principal and I is the cost of debt, as interest). FCFE is the cash flow available for equityholders. Income analysis of 40 firms sample in the 2008 shows that S varies from a minimum of € 1,654,000 to a maximum of € 52,177,000, having that the average sales are € 13,093,000. In the sample, 32 firms generate profits (Π>0) and 8 generate losses (Π <0); The average ratio Π/S in the sample is 3.62% with a minimum net income/sales ratio of -17.46% and a maximum value of 13.21%, the average Roe of the sample is equal to 2.34%, with a minimum value of -27.09% and a maximum value of 23.83%. The average Roa of the sample is equal to 10.98% with a minimum value of -0.06% and a maximum value of 12.31%. The average Ros in the sample is equal to 12.18% with a minimum value of -0.10% and a maximum value of 17.91%. The average capital turnover in the sample is equal to 0.721, with a minimum value of 0.166 and a maximum value of 1.221, the sample firms are characterized by high capital intensity (as turnover <1). Average ICR is equal to 121.90% with a minimum of -1.62% and a maximum of 560.51%. The analysis of the balance sheet shows an average TA of EUR 25.781 million to 2.861 minimum and € 68.214 maximum, average αWC_t / TA is 63,21% and FA / TA è 36,39%; αWC_i / TA is 41,96%. The other components of working capital (αWC_c + αWC_{ar}) / TA, is 21.25%. The analysis around the sources of capital in the sample shows, on average, that L is 5.314 and DER is 4.314 having that in the sample, on average, firms use high debt level to finance their investments so that D = 4.314E. In order to quantify the sources of liquidity, analyzing the creation of cash flow, data show a situation where average CF is 1.512 million euro, no negative value of CF, OCF average is 0.211 million euro, 11 cases of OCF negative on 40 firms; the average UFCF is -1.430 million €, with 19 cases of negative UFCF on 40 firms; average FCFE is -1.582 million €, with 23 cases of negative FCFE on 40 firms. The analysis shows that the sample firms have difficulty in generating a positive cash flow to serve debt (UFCF) and to distribute dividends to shareholders (FCFE). In particular, working capital has a substantial effect on the absorption of liquidity (0 cases of negative CF and 11 cases of negative OCF).

VII – Conclusions

Parma PDO Ham is the main product of cold cuts designation of origin in Italy at the level of production. The pork meat processing sector assumes importance in the economy of the province of Parma. The animals preserves sector shows that the turnover generated in the province of Parma, in 2008, is equal to 1,900 million euro, as 25.3% of sales in the food industry and 9.3% of the industry in general. In total of the Parma province, in respect of the meat processing industry, are involved 446 firms and others 143 local units, as for a total of 589 productive units, operate in the local industry, activating directly a total of 4,721 worker. Among the municipalities in the province, Langhirano, Lesignano de' Bagni, Sala Baganza and Felino could be considered territories with an increased level of activity. In those municipalities, with regard to the Parma PDO Ham, are concentrated 71.27% of firm and 80.28% of Parma PDO Ham production. Indeed, firms in the sector are characterized by significant fixed capital investment which should add significant capital equipment needed to support the cycle of maturing along with the long delays of payment required by large retail chains. So the analysis carried out on the sample of firms included in the study shows that there is difficulty in creating financial flows (UFCF) sufficient to support debt service (DS); in fact only firms with high size level of turnover, or production differentiation (as production of Parma Grosso) are able to generate not only profit for the shareholders, but also cash to ensure the sustainability of the business cycle, and the ability to distribute dividends or profits reinvestment to ensure discretionary investments (FCFE).

It emerges therefore that firms adopting strategies aiming to improve technological process to reduce production cost, even increasing profitability and positive cash flow generating capacity to support debt service and to reward equityholders. In addition, higher profit margins can be guaranteed by product differentiation (Parma Grosso) even having a greater focus on the goodwill of the product to the consumer by improving annexed services to the good, as the pre-slicing service that has been shown to have large spaces both on domestic and foreign markets.

Acknowledgements

The authors wish to acknowledge Camera di Commercio Industria, Agricoltura e Artigianato (Parma), Consorzio del Prosciutto di Parma, Unione Parmense degli Industriali (Parma), Istituto Parma Qualità (Parma), for the useful assistance and data, Paolo Camanzi (analisiaziendale.it) for the support in computational process, Elisa Manghi (University of Parma) for data collecting.

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The Nebrodi Black Pig: socio-economic analysis and perspectives (opportunities) of development

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Abstract. In recent years, breeding of Nebrodi Black Pigs is taking increasingly an own identity, abandoning that secondary role which saw it as income support and use of marginal areas of the farm. This push to production is due to increased attention and request that the consumer gives to local typical products of excellence. Although registered farms in ANAS are only 36, they are estimated at least a hundred. An exploratory research was conducted, then, on a sample of 36 farms. It provided the first socio-economic data (50% owned firms, average age of 49 years and 83% upper-middle level of education, etc.), farm size and territory (75% farms situated in the mountain, and 95% of those with wooded areas, etc.), end farm management (management of mating and birth, born pigs and period of weaning, feeding and veterinary care, etc.). Besides, results concerning the different farm productions, both farmers (annual production of pigs) and farmers-processors (type of processing and marketing), which represent the 33% of the farm sample under investigation and obtain a better economic performance, are reported. Finally, strategies that farmers hope to apply at the market for better utilization of local typical products and territory development are shown.

Keywords. Nebrodi Black Pig – Socio-economic analysis – Territory development.

Le Porc Noir des Nebrodi : Analyse socio-économique et perspectives de développement

Résumé. *Pendant ces dernières années, l'élevage des porcs noirs des Nebrodi a assumé de plus en plus sa propre identité, en abandonnant le rôle secondaire qui le voyait comme un soutien du revenu et comme une utilisation des zones marginales de l'exploitation. Ce moteur de la production est dû à une plus grande attention et une demande accrue des consommateurs concernant les produits d'excellence du terroir. Bien que les entreprises enregistrées dans l'Aire N.S.A. ne soient que de 36, on en a estimé au moins une centaine. Une enquête a donc été effectuée, sur un échantillon de 36 élevages qui ont fourni les premières données d'ordre socio-économique (50% des entreprises sont de la propriété de l'éleveur, qui a un âge de 49 ans en moyenne et parmi lesquels 83% ont un niveau d'instruction moyen-supérieur, etc.); de dimensionnement des exploitations et du territoire (75% des exploitations sont situées en montagnes et 95% de celles-ci en zones boisées, etc.), et de gestion de l'exploitation (gestion de l'accouplement et de la naissance, porcelets nés, période de sevrage, alimentation, soins vétérinaires, etc.). Le travail rapporte également les résultats concernant les différents produits de la ferme, à la fois pour les éleveurs (production annuelle de porcs) et pour les éleveurs-transformateurs (type de transformation et de commercialisation), qui représentent 33% de l'échantillon des entreprises sous enquête et obtiennent un meilleur résultat économique. Enfin, on montre les stratégies que les agriculteurs souhaiteraient appliquer sur le marché pour une meilleure utilisation des produits du terroir et pour le développement local.*

***Mots-clés**. Porc Noir des Nebrodi – Analyse socio-économique – Développement local.*

I – Introduction

The ancient civilization of peasants and shepherds of Nebrodi is reflected in numerous handicrafts. The food products find their highest expression in dairy products as the canestrato cheese, the pecorino cheese, the provola cheese and the ricotta cheese and the famous cold meats made with meat from Nebrodi Black Pig. In recent decades the influence of globalization on animal production systems has led to a loss of biodiversity. From this awareness society has moved to a rediscovery of ancient production systems of the indigenous breeds considering the influences of the environment in which they have developed and adapted.

The Nebrodi Black Pig, added to the list of native species in danger of extinction, has very ancient origins and until recently was only a supplementary income business. Marginal areas, not otherwise used as pasture or arable areas, represented by the forests and the Mediterranean scrub, were the primary source of food. The pigs belonging to this breed live in the wild in the densest and most inaccessible woods of mountainous areas of Nebrodi, where they reproduce in their natural state, and because of their characteristics are well adapted to harsh environmental conditions of the territory, where other races would have difficulties to adapt.

The Nebrodi Park, established in 1993, represents, together with the Park of Etna, Alcantara and Madonie, the largest protected area of Sicily with its 85,587 ha, and includes the most important and extensive woodlands in Sicily (Fig. 1). Half of the woodland consists of coppice, and the other by tall trees. The most important tree species are represented by *Fagus sylvatica* (the extreme southern limit of diffusion area), *Quercus cerris* and *Quercus suber*. There are also some formations of *Quercus ilex*, *Taxus baccata, Ilex aquifolium* and important lacustrine and rocky environments.

The livestock system of Nebrodi is characterized by mixed farming (cattle, sheep, goats, horses, pigs, etc.) bred in the wild, native species, inaccessible terrain, lack of roads, common grazing and transhumance in some periods of the year. In this context, livestock, agricultural and health management are difficult to implement. In addition, the "livestock Nebrodi system" is a natural and unique symbiosis of factors in which ancient indigenous germplasm, environment and type of nutrition are the basis for the production of excellent local products highly demanded by consumers.

The preciousness and uniqueness of the meat and the typical and natural products derived from the Nebrodi black pig, recognized by consumers, has led by about twenty years, some farmers to change the type of breeding black pigs. Favouring this rise in the market, and in order to increase the number of animals bred and to better organize the chain of production, they tried to optimize the outdoor rearing, using the forest areas at farms' disposal and intervening in feeding by additions during periods of shortage of natural foods. This type of farming evokes the concept of animal welfare and is in line with consumers' demand for those products derived from "stress free" animals. Although the black pig has a slow growth compared to other commercial breeds, the organoleptic characteristics of its meat and fat, used to produce processed meats such as hams and cold meats, are particularly appreciated by consumers.

The products of the forest, in fact, give the meat of these animals qualities of high value, and the increased interest and awareness of farmers to their genetic heritage, has meant that the black pig became a slow food presidium and for its fresh meat was asked the DOP award. Although we said above, the farms registered in the "National Swine Breeders Association" (ANAS), in 2009, appear to be about 40, but they are estimated to be at least a hundred in the territory of Nebrodi (Fig. 2). As shown in Fig. 2, the black pig farming is mainly located in the province of Messina (61.5%). We also remark that in the province of Benevento, there is the presence of a group of Nebrodi black pigs, exclusively for research purposes, belonging to the Consortium for the Testing, Spreading and Application of Innovative Bio-techniques (CONSDABI). The purpose of this survey is to provide information about socio-economic holdings of the Nebrodi Black Pig from a perspective of development of this zootechnic sector.

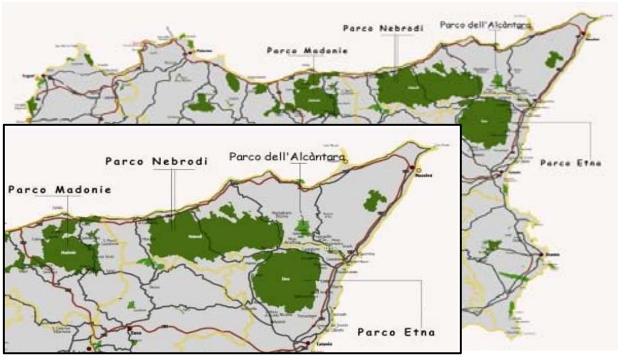

Fig. 1. Parks in Sicily.

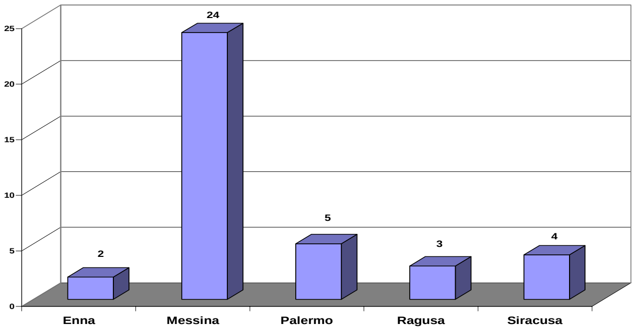

Fig. 2. Diffusion of farms of Nebrodi's Black Pig in Sicily.

II – Materials and methods

The survey involves 36 farms of Nebrodi's Black Pig located in municipalities of Sinagra, Mirto, Militello Rosmarino, S.Fratello, Longi, Frazzanò, Ucria, Castell'Umberto, Tortorici, Alcara Li Fusi, Naso, Caronia, Floresta, Capizzi and S. Lucia del Mela. A large part of them are located within the protected area of the Nebrodi Park (Fig. 3).

Fig. 3. Georeference of farms and area of Nebrodi Park.

The recognition of socio-economic aspects of the sample involved was obtained through an interview to the Nebrodi black pig farmers.

The structured interview consists of the following areas of investigation:

(i) Data about the breeder (the kind of farming management: property, tenancy and mixed; age and school level).

(ii) Data on companies and territory (size, location and presence of wooded areas and / or pasture).

(iii) Data about farm management (pig shelters, management of reproduction, age at the time of slaughter, feeding, medical support, production and marketing).

(iv) Data about the needs / demands of the farmers (enhancement of products and technical assistance).

III – Results

1. Data on farmers

The details obtained show that the sample farms are 50% owned by the farmers themselves, that their average age is 45.3 years about and that 83% of them have a middle or upper school education level (Fig. 4). From these data it would seem that in this area, the age may be considered an indicator of greater experience and accountability, arousing an interest to undertake the breeding of Nebrodi Black Pigs, in people having a fairly good school level.

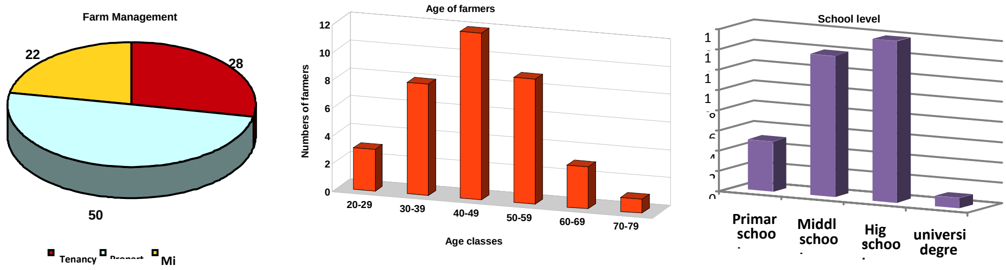

Fig. 4. Distribution of farms by management, age and schooling.

2. Data on the farms and the territory

The analysis shows that 50% of farms are small dimension, localized in the mountains and characterized by the presence of woodland and pastures (Table 1).

The distribution of vegetation in the territory of Nebrodi, includes hills with oaks, chestnut trees and Mediterranean scrub, and mountain areas where, in addition to previous species, there are also turkey oak, beech and holm-oak. In grazing areas, the predominant species are composed largely of wooded and arable areas with prevalence of hay, oats, vetch, barley and field beans, that are used in the farm. The sample of 36 farms is formed as following: 10 farms have only forest areas, 3 farms only grazing areas and 23 farms both forest and grazing areas. Farms with forest areas in their territory are 33 (91.6%: 23 with pastures and 10 with just forest areas). In the 23 farms with woods and pastures has been analyzed the distribution of the prevalence of these areas, noting that for most farms have forest areas (Table 2). Concerning the extension of the surface, small farms with wooded areas (73%) prevail. Finally, farms having pastures in their territory are 26 (23 with forests and 3 with pastures only). There are many farms having not very extensive pastures.

Table 1. Distribution of farms by size, location and presence of forests and pastures.

Farm size	%	Slope	%	Presence of wooded areas and/or pasture	%
1 - 25 ha	50	Mountain	75	Farm with wood and pasture	64
26 - 50 ha	22	Hill	16	Farm with only wood	28
51 - 75 ha	6	Plain	6	Farm with only pasture	8
>75	22				

Table 2. Prevalence and size of farms with forests and pastures

Farms with mixed areas	
Prevalence of forest	40%
Equity between forest and pasture	30%
Prevalence of pasture	6%
Farms with wood for area classes	
1-25 ha	73
> 25 ha	27
Farms with pastures for area classes	
1-25 ha	73
> 25 ha	27

3. Farm management

The results show that more than half of the farms provides farrowing sows with the presence of shelters or even with specific pig shelters called "Zimme", and in most cases there is not any programmed mating. The litter occurs mainly indoors in individual pens, but there are also farms in which the parturition occurs outside (Fig. 5).

The number of piglets born for each sow is about 9, but the mortality percentage observed is around 16%, especially in farms where the sows litter occurs outside. Usually piglets are weaned between the 35th and the 60th day, and the average reproductive life of sows is 5.4 years (Table 3).

The scatter plot (Fig. 6) shows that the modal value on the slaughter age is about 12 months, and that the weight of the pig is about 70-110 kg., depending on the type of farming and on the type of nutrition.

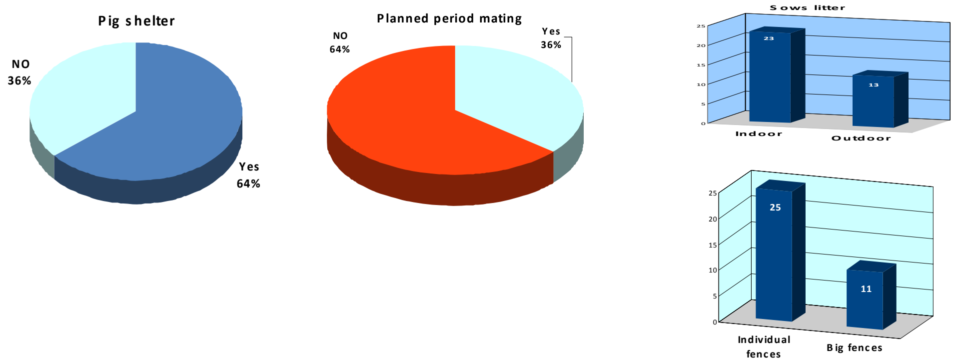

Fig. 7. Presence of pig shelters, mating and parturition

Table 3. Number of piglets born and mortality, age of weaning

Variable	Average	Mode	SD
Piglets born/sow	8.7	8	0.24
Pigletys dealth rate (%)	15.97	10	1.6
Sow slaughtering (years)	5.4	5	2.46
	No. of farms	**%**	
Age of weaning			
Between 30 and 50 days	14	39	
About 60 days	14	39	
Between 90 and 120 days	5	14	
No weaning	3	8	

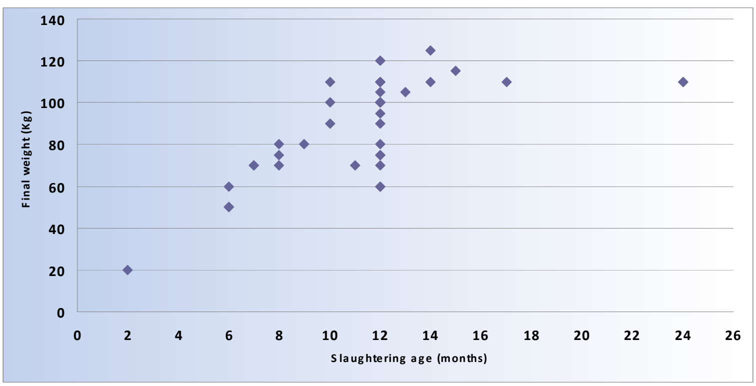

Fig. 6. Age of slaughter and final weight.

4. Feeding system

The composition and origin of food is varied and depends on the availability of the farm, on forest areas destined for the use of pasture, on different periods of the year, and on the years with higher or lower production of natural food in the forest. All farms having forest areas at their disposal allow to the animals to graze with the products of the forest and undergrowth. When no food is available for grazing (in winter with snow-covered land, or in summer), pigs are fed on cereals and cereal-based commercial feed. The natural diet of pasture consists mainly of acorns, chestnuts, and products of the Mediterranean scrub. When the integration is realized, it is composed by grains such as barley, germinated barley, field beans, bran, corn and grain. These are given either whole or ground, depending on availability of millstones in the farm. The commercial foods, specially formulated and balanced by category, are composed by mixed cereals and can be both ground and pellets. There is a small percentage of farms, which create their own composition of feed always based on field bean, barley, wheat, corn, carob, bran, supplemented with minerals.

5. Veterinary support

Only one farm uses vaccination against colibacillosis for mortality in piglets, and two carry out preventive vaccination against Aujeszky's disease.

The hardiness, the resistance against diseases and adverse climatic conditions, the state of extensive breeding allow almost all farms do not have serious health problems excluding skin, lung and gastrointestinal parasitic infestations, and rare infectious problems to respiratory system.

6. Production and marketing

The production activity of the farms is 47.5% only directed to breeding with sale of weaned piglets or pigs at the end of the finishing stage. While 8.3% of the farms carries out both breeding farm and processing.

The marketing is carried out following different models:

(i) "Long-chain":

Breeder -> traders of animals -> slaughter -> butchers -> seller of fresh product at retail.
In this case the farmer does not address the handling of animals in first person.

Breeder -> slaughter -> butchers -> seller of fresh product at retail.

In this case the farmer addresses the handling of animals and sells them.

(ii) "Medium chain":

Breeder - transformer -> slaughter <-> farmer - processor -> distribution.
In this case the transformer - farmer relies on other professional figures in the marketing and distribution.

(iii) "Short chain":

Breeder - transformer -> slaughter <-> breeder - transformer - the seller
In this case the farmer, being transformer, becomes also a seller of his products.

However, some of these transformers are also sellers of their products in accommodation structures (restaurants, specialty shops with tasting, etc.)

With regard to the revenues obtained, the 69.4% represented by the long-medium chains' model, believes that it is not sufficient or just sufficient, while the 27.7%, represented by the short chains' model, is quite satisfied with own economic productivity.

7. Demands of farmers

Strategies for better enhancement of products. The near unanimity of the interviewed farmers think that is important the creation of a PDO and to be membership of a consortium for development. All respondents agree on the request of a better publicity, diffusion and knowledge of Nebrodi black pig, both at regional and national field. They remark also the need to inform the consumer about the organoleptic characteristics of the products and about high quality of the raw materials used, in order to improve the marketing through a widespread distribution.

Technical assistance. About 72% of farmers would like to have more assistance in the management of breeding, specifically concerning the quality and the quantity of food, the decrease of mortality, in addition to a better health care. 10% also requires technical assistance to improve and refine their processing techniques of the products.

IV – Conclusions

This survey is a description of the of Nebrodi black pig breeding provided by the farmers themselves. The farming system used is mainly a semi-wild type by using the forest resources more in finishing period. The breeding of the Nebrodi Black Pig is a closed-cycle type. The most of respondents is only responsible of the breeding (88%) and relies to other professional figures to the processing and sale of products. It is clear the importance of a suitable health management, that would allow the increase in productivity and would ensure to obtain safe products for the protection of consumers' health, as required by the farmers themselves.

The awareness about the preciousness and uniqueness of the meat and products derived from natural and typical Nebrodi black pig, is associated with the firm conviction that the various phases of the production chain can not be approximately carried out. In this regard, the farmers interviewed emphasize this need due to their awareness about a lack of their organizational structure. In addition, the demand for environmental quality by citizens continues to grow and is becoming even more qualified and functional. In fact, the citizen, as a consumer, adapts its behaviour concerning rural tourism and gourmet or the purchase of agricultural food and products or handicrafts, on the bases of the information received about the environmental quality of territories. European consumers also require high quality food products made using production systems compatible with environmental, landscape and genetic resources conservation. In this regard, traditional foods produced by local farms and traditional practices, that can help to achieve these goals, can have significant effects on innovation and added value for rural societies. In conclusion, the breeding of Nebrodi black pig, is an opportunity for regional development through environmental protection, soil conservation and preservation of biodiversity. In summary, through an innovation, respectful of tradition, young generation can be encouraged limiting the unstoppable phenomenon of rural exodus.

Acknowledgements

The authors wish to acknowledge the financial support received from the Programme MED / MED Programme 2007-2013 / QUBIC Animal Breeding: Quality Biodiversity Innovation Competitiveness, Sicily Region - Department of Agriculture.

Thanks for technical assistance to: E. Rappazzo, G. Rubino, G. Romeo, A. Lastra, S. Bellinvia.

Pig breeders in extensive systems based on local breeds: Stakes of their insertion in the development of the territories

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Abstract. Within the Mediterranean QUBIC project centered on innovations developing biodiversity, we approach the points of view of the pig breeders in extensive systems based on local breeds, on their insertion in the dynamics of territorial development. A questionnaire including 4 parts and 12 questions was managed towards 123 farmers carrying on their activities in the 5 areas interested by the project: Italy (Tuscany, Sicily and Emilia-Romagna), Greece (Thessaly) and France (Corsica). Data collected relate to (i) environmental problems, (ii) visions of the territory and local insertion of activities, (iii) local breed seen as a factor of anchorage of the activities, and (iv) professional identity of the stockbreeder of local breed. We carried out an analysis centered on the link between breeds and territory, in order to identify possible points of blocking and levers in the projects of development of these breeds. We identify common features but also marked differences: (i) established systems (Cinta Senese in Tuscany) where a lot of newcomers show some lack of technical culture; (ii) stabilizing systems (Nustrale in Corsica, Nero Siciliano in Sicily) with deep anchorage of activities but weak professional organization; and (iii) emerging systems (Greek in Thessaly, Nero di Parma or Romagnola in Emilia-Romagna) not yet insured in their territorial insertion. Such comparative study allows supplying useful elements for future exchanges at Mediterranean level.

Keywords. Pig – Breeders – Local development – Extensive systems.

Les éleveurs porcins en systèmes extensifs basés sur des races locales. Enjeux de leur insertion dans le développement des territoires

Résumé. *Dans le projet méditerranéen QUBIC centré sur les innovations valorisant la biodiversité, nous approchons les points de vue des éleveurs porcins en systèmes extensifs basés sur des races locales, sur leur insertion dans le développement territorial. Un questionnaire comprenant 4 parties et 12 questions est administré auprès de 123 éleveurs dans les 5 régions du projet : Italie (Toscane, Sicile, Emilie-Romagne), Grèce (Thessalie) et France (Corse). Les données recueillies portent sur (i) les problèmes environnementaux, (ii) les visions du territoire et de l'insertion locale, (iii) la race locale comme ancrage des activités et (iv) l'identité professionnelle de l'éleveur. Nous effectuons une analyse centrée sur le rapport des races au territoire afin d'identifier des points de blocage éventuels et des leviers dans les projets de développement territorial. Une analyse comparative dégage des traits communs mais aussi des différences marquées :(i) des systèmes établis (Cinta Senese en Toscane), où un grand nombre de nouveaux éleveurs montrent un manque de culture technique ; (ii) des systèmes en cours de stabilisation (Nustrale en Corse ou Nero Siciliano en Sicile), avec un fort ancrage territorial mais des faiblesses organisationnelles ; et (iii) des systèmes émergents (Grec en Thessalie et Nero di Parma ou Romagnola en Emilie-Romagne), dont l'insertion territoriale demeure très incertaine. De tels travaux permettent de disposer d'éléments utiles pour de futurs échanges au niveau méditerranéen.*

Mots-clés. *Porc – Éleveurs – Développement local – Systèmes extensifs.*

I – Introduction

Give the floor to the breeders' voice, such intention is not so frequent in our scientific communities (Flamant *et al.*, 1994). What about the breeders' point of view on some main questions we, as scientists, are studying?

The work takes place within the framework of the Mediterranean project centered on the innovations to develop biodiversity (QUBIC): Animal breeding - Quality Biodiversity Innovation Competitiveness. Is that biodiversity good for the future of the production units as Iberian pig is demonstrating (López-Bote, 1998)? Is the local breed an asset at territorial level?

A questionnaire including 4 parts and 38 questions was managed towards 123 farmers carrying on their activities in the 5 areas interested by the project: Italy (Tuscany, Sicily and Emilia-Romagna), Greece (Thessaly) and France (Corsica). In each area, there is one local breed, excepted in Emilia-Romagna where there are two. So, local pig breeds considered in our study are :

- ✓ For Italy, in Tuscany the Cinta senese breed, in Sicily the Nero Siciliano breed, in Emilia-Romagna both the Mora Romagnola breed and the Nero di Parma breed.
- ✓ For Greece, in Thessaly, the Greek breed.
- ✓ For France, in Corsica, the Nustrale breed.

We decided to focus only on some of these data in order to present a more accurate analysis on the specific linkage between local breed and territory according to the breeders' point of view.

II – Material and methods

1. Breeders sampling

As reported in Table 1, the sample of breeders interviewed is quite important with 123 farmers in 5 of the areas of the project.

So these 6 local pig breeds are comparable as census of animals is quite reduced and breeds are still located in the native area.

Table 1. Sample of breeders according to the country, the region and the breed

Country	Italy	Italy	Italy	Greece	France
Region	Tuscany	Sicily	Emilia-Romagna	Thessaly	Corsica
Name of the breed	Cinta Senese	Nero Siciliano	Mora Romagnola and Nero di Parma	Greek	Nustrale
Number of interviewed breeders	35	36	19	10	23

2. Data recollected

A questionnaire including 4 parts and 38 questions was managed towards the 123 farmers on the following fields:

- Part 1: Environmental problems and the way the farmers face them.
With a total of 8 questions, only 3 questions are presented.

- Part 2: Visions of the territory and the insertion of breeding activities in the local dynamics.
A total of 10 questions and 2 questions presented.

- Part 3: The local breed seen as a factor of anchorage of the activities.
On 10 questions, 5 questions are presented.

- Part 4: The professional identity of the stockbreeder of local breed in the evolutions of the sector.
Among 10 questions, 3 questions are presented.

Results are expressed as % of YES according to the question.

III – Results

According to answers, we carried out an analysis field by field. Such analysis is centered on the link between the breeds and their territory within these various located systems, in order to identify possible points of blocking and levers in the projects of territorial development of these breeds.

1. Environmental problems

In the first part of our enquiry, we are dealing with environmental problems and the way the farmers face them. In particular, questions of pollution and sanitary risks of the animals in free range.

Q 1 – Are you facing some environmental problems in your livestock farming? Which ones? (For example water pollution, soil erosion, plants and trees destruction, animal divagation)

Q 2 – What kinds of disease are present?

Q 3 – Due to extensive livestock system, are you obliged to consider wild animal diseases in your prevention plan?

Table 2. Answers from the breeders about environmental problems

Answers from the breeders	Cinta Senese	Nero Siciliano	Mora Romagnola Nero di Parma	Greek	Nustrale
Q1	60	0	58	0	56
Q2	Pneumony, Parasites	Parasites	None	None	Aujeszky
Q3	18	0	5	10	0

The grazing in forest seems to be commonly used and almost a great part of the pigs' life is outdoors. Nevertheless, a majority of the breeders are declaring no environmental problems, but we notice great differences between the various situations.

Parasites are clearly the main kind of disease as mentioned in the answers.

Soil erosion and some trees destruction are also evoked but breeders are complaining to the obligation to put nose ring for avoiding such problems.

And very few breeders have consciousness of the questions of contamination from the wild animals (especially from the wild boars).

As major issue, we can see that the great part of breeders is not aware of environmental problems.

2. Insertion within the territory

In the second part of our enquiry, we are looking for the visions of the territory expressed by the breeders and the insertion of their activities in the local dynamics. In particular, we try to approach organizational aspects of the breeds' management and of the product valorization in the territory and the factors of specificity of the extensive breeding in the offer of regional products.

Q 4 – Do you think breed goodwill and territory linkages represent added value for you?

Q 5 – Is the breed well known in the territory and do you use the image of the breed and/or of the territory to sell your products?

Table 3. Answers from the breeders about their insertion within the territory

Answers from the breeders	Cinta Senese	Nero Siciliano	Mora Romagnola Nero di Parma	Greek	Nustrale
Q4	100	100	95	60	100
Q5	95	100	74	70	74

This second part of enquiry is giving less contrast between the various situations.

In general, the local breed is conferring good insertion to the breeder and positive image for its activity.

In addition, some lack of recognition at social level is mentioned by breeders (excepted in Sicily) and the local breed is no sufficient to insure a good position in the local society.

In Corsica, breeders are also mentioning risks of confusion at market level, as products deriving from the local breed have no special identification at the moment.

As main issue, we can assume that the image of the breed is adding value on an effective way. And the breeders are using this image for commercial purposes.

3. The local breed as an asset

This third part of the enquiry deals with the local breed seen as a factor of anchorage of the activities. The adequacy of the animals to the systems of breeding and the collective management of the breed are the principal points as well as the question of the possible crossbreeding with other selected races.

Q 6 - Do you feel that the local breed is insuring you a deep anchorage in the territory?

Q 7 - Do you consider the local breed as fully adapted to the local farming system?

Q 8 - Do you establish a link between the "good breeder" and the "beautiful animal"?

Q 9 - Have you effective practices of crossbreeding?

In this third part, we can see a real consensus for the questions 6 and 7 as quite all the breeders are considering deep anchorage provided by the local breed and also a good adaptation of their animals to the local farming system.

For the questions 8 and 9, we notice great contrasts among the various situations.

For Cinta senese and Mora Romagnola / Nero di Parma, the crossbreeding is an ancient practice and breeders are considering it without any problem. A special name is given for crossbred animals in Emilia-Romagna. In Greece, as recovering of black pigs is still in progress, crossbred animals are quite the normal situation. In the other areas, such as Sicily and Corsica, this practice is disappearing moving to the pure local breed as a main stream.

Table 4. Answers from the breeders about the local breed and the crossbreeding

Answers from the breeders	Cinta Senese	Nero Siciliano	Mora Romagnola and Nero di Parma	Greek	Nustrale
Q6	100	95	74	60	83
Q7	100	95	90	100	74
Q8	90	10	79	100	17
Q9	36	26	55	50	21
Main issues	Crossbreeding as ancient practice	Rejection of “good breeder”	Crossbreeding = “Borghigiano”	Crossbreeding considered as normal	Rejection of “good breeder”

For Corsican and Sicilian breeders, they reject the notion of “good breeder” and they consider that local breed animals can be diverse according to the breeders’ preferences. They assume an internal diversity as breed collective identity.

As main issue, local breed insures deep anchorage to the territory and seems to be well adapted.

4. The local breed as a professional identity basis

In the last part of interviews, we emphasize the professional identity of the stockbreeder of local breed in the evolutions of the sector. Information is in particular collected on the anteriority of the breeding activity, their vision of their trend compared with other types of breeding, as well as the pride to be a producer of local breed.

Q 10 – Are you claiming to be considered as a distinguished activity compared to exogenous breed farmers?

Q 11 – Is the local breed a familial heritage transmitted by the previous generations (not something completely new)?

Q 12 – Are you feeling proud to be a local breed promoter?

Table 5. Answers from the breeders on the professional identity

Answers from the breeders	Cinta Senese	Nero Siciliano	Mora Romagnola Nero di Parma	Greek	Nustrale
Q10	100	100	84	30	30
Q11	14	100	10	40	87
Q12	100	95	68	70	74

Professional identity of the breeders seems to be a crucial point for their territorial insertion. As a majority of breeders is claiming to be differentiated from the exogenous breed farmers, the national situation must be taken into consideration: in Italy, the local pig breeds are giving a strong identity to the farmers.

We notice a lot of newcomers, in Cinta Senese, Mora Romagnola and Nero di Parma, and quite important in Greek. At the contrary, Corsican and Sicilian seems to have mainly familial heritage.

Quite all the breeders are associating pride and local breed activity. This point is very important to be underlined because the future of the local breeds could be facilitated by such a feeling.

As main issue, major part of breeders are proud, but are also claiming to be better recognized.

IV – Discussion

A comparative analysis makes it possible to identify common features but also marked differences.

As common features, we can identify a low perception of environmental problems by quite all the breeders. Even if the risk of damage to the natural resources is obvious, it seems to be of minor interest in breeders' point of view. This point must become a priority for the extension services in order to avoid further disqualification of outdoor systems for pig production.

The local breed is conferring a deep anchorage to farmers and animals are considered as well adapted to the way of rearing. The image of the local breed is giving some advantage to the breeders and they use such image when marketing the products (Casabianca and Fallola, 1994). All the breeders seem to be proud to be local breed promoters but they are claiming for a better recognition at social level.

As main contrasts, we must distinguish the trajectory of each situation. The evolution of the breed is giving an orientation to the whole sector. We identify:

(i) Established systems such as *Cinta Senese* in Tuscany. With a PDO already recognized at national level, the breed seems to be clearly stabilized. But we notice a lot of newcomers attracted by the reputation of the breed and a lack of knowledge and technical culture in the management of outdoor systems.

(ii) Stabilizing systems as the *Nustrale* in Corsica or the *Nero Siciliano* in Sicily. Both of them are deeply rooted in local culture and applying for a PDO inducing some new questions (Lambert-Derkimba *et al*., 2011). Breeders show a family heritage and the technical culture is enforced by generations.

(iii) Emerging systems as the *Greek* pig in Thessaly and the *Nero di Parma* or the *Mora Romagnola* en Emilia-Romagna. In such situations, professional organizations are still lacking to ensure the future of the breed.

This type of interviews analysis, mixing qualitative and quantitative data, is useful to identify key topics and stakes for the breeders themselves and for territorial management of local breeds. This type of analysis also allows wide comparative study at a Mediterranean scale.

Those key topics should be further studied thanks to a more qualitative analysis, using for instance semi structured interviews. Such a qualitative analysis should allow gathering breeders' discourse trying to minimize the influence on the orientation of the answers, without a pre-construction of the themes. It could allow understanding better the dynamics of territorial management for local breeds and the stakes on different territories, and complementing usefully this first comparative approach.

V – Conclusion

According the differences we identified, we carried out a “gap analysis” centered on the link between the breeds and their territory within the various systems, in order to identify possible points of blocking and levers in the projects of territorial development of these breeds.

Some clear conclusions can be provided for each type of situation and breed.

For Cinta Senese : The situation is characterized by a lot of newcomers without experience, and some environmental problems not really taken into account by the PDO specification. The processed products should be protected in addition of fresh meat, because of risks of confusion at market level. It should be interesting to compare this situation to the Iberian pig situation where PDO is obtained for a long time.

For Nero Siciliano and Nustrale : Some similarities are observed between the two islands as this activity is rooted in the local culture. But farmers seem to face great difficulties to innovate and to organize. Environmental issues should be emphasized and PDO protection should be completed.

For Mora Romagnola, Nero di Parma and Greek : The three breeds are not really ensured till now. Breeders show a weak situation to be reinforced mainly at organizational level.

Such comparative studies based upon breeders view points are supplying useful elements for future exchanges at Mediterranean level.

Acknowledgments

We thank the QUBIC project for giving us the possibility to manage the same questionnaire in the several areas of the project. We also want to thank all the scientific and technical staff from the various project partners involved into the data gathering and obviously the breeders willing to answer to our questions.

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Iberian pig price cycle: Extremadura market

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Abstract. This work analyses the pattern of Iberian pig prices at the Extremadura market, in order to find out about the cyclical fluctuations of Iberian pig prices, both for piglets and fattening animals in their different commercial categories: "Bellota", "Recebo" and "Cebo". The study looked at prices between 1989 and 2009, using the monthly average prices for those categories at the Extremadura market, and deflating them by the Producer Price Index. These deflated values enabled us to set 12-month Centered Moving Averages to offset the product's seasonal nature. The prices went through three cycles during the research period. The cycle for the piglet category was seen to fluctuate between 41 and 86 months, with an average of 74.8, and the average duration of the cycle for fattening pigs of the "Cebo" category was found to be similar (between 43 and 87 months, with an average of 74.4 months). A seasonal pattern was observed in the prices for the piglet category, with minimum prices in summer and maximum prices in winter, whereas the fattening animals reached their minimum prices in autumn and maximum prices in winter.

Keywords. Iberian pig – Prices – Cycles.

Cycle de prix du porc Ibérique : Marché de l'Estrémadure

Résumé. *Cet article analyse l'évolution des cours du porc Ibérique dans les halles d'Extremadura, tant pour les porcelets que pour les porcs engraissés, correspondant respectivement aux différentes qualités commerciales de "Bellota", "Recebo" et "Cebo", afin de suivre les variations cycliques des cours. La période d'étude porte sur les années 1989 à 2009. Les cours mensuels moyens par catégorie aux halles d'Extremadura, corrigés en fonction de l'indice de prix industriels, ont été utilisés. Ces cours mensuels moyens corrigés ont été utilisés pour déterminer les moyennes mobiles centrées de 12 mois, afin d'éviter les variations saisonnières. Dans la période d'étude il y a trois cycles pour ces cours. Le cycle de la catégorie porcelets oscille entre 41 et 86 mois, avec une moyenne de 74,8. De même la longueur moyenne du cycle des porcs engraissés est de 43 à 87 mois, avec une moyenne de 74,4 mois. Une certaine saisonnalité des cours est observée pour les porcelets, avec des prix minimaux l'été et maximaux l'hiver; et pour les animaux engraissés, avec des valeurs minimales l'automne et maximales l'hiver.*

Mots-clés. *Porc Ibérique – Prix – Cycles.*

I – Introduction

Pig production in Spain is mainly based on the intensive production of improved white-coat pigs. However, there is also an extensive production based on the Iberian pig, both pure and cross-bred with Duroc Jersey.

In recent decades, this indigenous race has undergone a period of decline, particularly between the 1960s and the 1980s. This has been caused by a number of factors, including zootechnical (lower prolificacy and growth rates than other improved races), social (diminishing agricultural population, leading to a crisis in traditional production methods), dietary (Spanish society has rejected meat that is high in fat), pathological (the Iberian pig was hit hard by African Swine Fever) and economic (other meat products, such as chicken or white pork, offered more competitive prices).

However, it has clearly been making a comeback in recent years. The decline that came about around 1993, as a result of lowering animal prices, was followed by a period of spectacular growth. Furthermore, the demand of Iberian pork products in the Spanish market was on the rise, with ever increasing appreciation of their quality. Consumers were willing to pay for that quality, as they now had greater spending power.

The marketing of the Iberian pig is characteristically varied and complex. This is due to the range of categories throughout the production cycle, to the races used (pure or cross-bred) and to the different types of feeding used in the final fattening stage: “Bellota” or Montanera (mainly acorns), “Recebo” (mixture of acorns and fodder) and “Cebo” (animals are exclusively fed compound feed, either as part of an intensive or extensive system).

The marketing of the Iberian pig has also been influenced by factors such as the modernisation of farms, which are today managed more rationally and competitively than before; the extended slaughter season, due to improvements in ways of preparing and preserving pork products; and the rise in the standard of living.

Situations in which there is a surplus or deficit in animal production, a shortage or abundance of acorns and pasture, demand for cured products, etc. mean that prices rise and fall on a fairly regular basis, much as they do for other products, and more specifically, in the case of the white-coat pig that is intensively farmed.

The pattern of prices and production of the white pig has been studied on a number of occasions (Zorrilla, 1969; Weinberg and Sobrino, 1958; Caldentey, 1967, 1980; De Pedro *et al.*, 1984; Caldentey and de Haro, 1985; Riopérez and Paz, 1986; Agote, 1991; Muñoz, 1998 and Berrocal, 1999; Dawson P.J., 2009) to discover how it adapts to different production forecasts over relatively wide periods of time.

In the case of the Iberian pig, however, its seasonal production and the irregularity of information about its prices have made it difficult to carry out a market analysis of this type of livestock (Berrocal, 1999, Muñoz, 1998), as well as the fact that only relatively recently has a record been kept of the weekly or monthly prices of the different types of animals sold in farming markets, which makes it difficult to have a clear view of price behaviour.

These circumstances, and the importance of Iberian pig production in south-west Spain, have created an interest in finding out more about the evolution of Iberian pig prices in the main area of pasture production.

The objective of this paper is to analyse the cycle of Iberian pig at the Extremadura market, which will enable us to find out about its pattern and so have an idea about price behaviour.

II – Material and methods

For this work we have used the average monthly prices for piglets and “Cebo” pigs at the Extremadura market, as these are the only categories of pig that have recorded prices all year round. The average prices were deflated with the Producer Price Index (PPI), with base year 2005, obtained from the [Spanish] National Statistics Institute, in order to counteract the effect of inflation in the period under analysis.

The prices’ seasonality was offset by means of the Centered Moving Averages of the monthly prices that was put forward by Caldentey (1980), applying the following formula:

$$X'_t = 1/24 * (X_{t-6} + 2X_{t-5} + \ldots + 2X_{t-1} + 2X_t + 2X_{t+1} + \ldots + 2X_{t+5} + X_{t+6})$$

Where t represents the months considered.

III – Results

Figure 1 shows the values of the centered moving averages for the piglet and “Cebo” pig prices at the Extremadura market, together with the pattern of the quarterly variations in GDP between 1989 and 2009. In this graph it can also be observed how both series of data behave in a similar way. With regard to the average duration of the two categories’ cycles, there are hardly any differences; in the case of piglets, this duration is 74.8 months, and in the case of “Cebo" pigs it is 74.4 months. In general, however, we observe no coincidence in the maximum and minimum prices for both series, and they also come about with no fixed pattern of differences. Espárrago *et al.* (1999), in a shorter period of time than in our study, also pointed out that the average duration of piglets and “Cebo” pigs was very similar, with an average cycle duration that was just 6 months shorter.

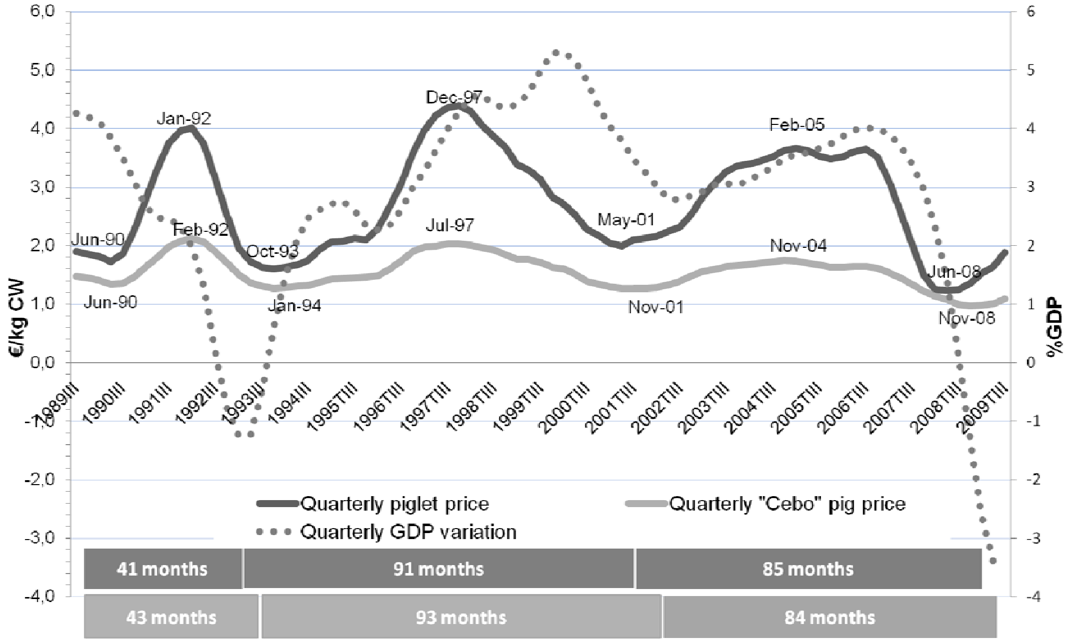

Fig. 1. Chart showing the centered moving averages for the quarterly deflated prices of piglets and Cebo pigs and the GDP variation between 1989 and 2009.

In Fig. 1 it is worth pointing out the size of the wave drawn by each cycle. We can clearly see here how, in the case of the piglets, these fluctuations are much more pronounced than in the case of the “Cebo” pigs, where the curve drawn by this series of data is much gentler, which could be due to the rigidity of the short-term supply of piglets, as described by De Pedro *et al.* (1984).

We can see how, for piglets, the price drops are very abrupt, whereas their price recoveries are much slower; however, if we look at the data for “Cebo” pigs, we observe that it is completely the opposite; this could be due to the different production process involved in each category. The need to sell piglets at a given weight would prevent them from being sold at a later point in time, whereas in the case of the “Cebo” pigs, the time at which they are slaughtered may be sooner or later, according to market prospects.

Furthermore, we would like to highlight the rapid fall in prices, both for piglets and “Cebo” pigs, in the period of time between January 1992 and October 1993, compared with the evolution that had come about in subsequent periods of time. We do not have enough information to know what cycles were like prior to June 1990, but in 1991, with the purpose of eradicating African Swine Fever (ASF) in Spain, ASF-“free”, “monitored” and “affected” zones were established ([Official State Gazette] BOE-A-1988-29631, 1988; BOE-A-1991-7267, 1991; BOE-A-1992-26539, 1992). Extremadura was in the affected zone, and was not declared a monitored zone until November 1993. It was not until a year later that it was finally declared an ASF-free zone. These limitations in the animal trade could be one of the causes of that heavy fall in prices in both markets, and the classification as a monitored zone in November 1993 could have been the reason behind the price recovery.

The standstill in 1995, when there were no longer any limitations on the trade of animals and Iberian pork products, coincided with periods of heavy drought and a halt in the GDP growth in Spain. Once this period had passed, the sector once again picked up the growth it had been undergoing in the previous cycle, even reaching higher values than before.

In the second and third cycles (January 1994-November 2001 and November 2001-November 2008, respectively) we can see that the price extension for “Cebo” pigs coincided with a period of high growth in the Spanish economy, which leads us to suppose that the extension of the second cycle was produced as a result of the high demand for animals by industrialists who were motivated by the good economic expectations of the time. This situation meant that the surplus supply that was produced in 1997 was not absorbed in full, as would be expected from looking at the cyclical behaviour of prices in preceding years. Instead, the surplus supply meant that, between December 2002 and October 2006, prices did not reach the maximum values observed in previous cycles, despite the fact that the positive economic situation in the country kept the “Cebo” pig prices high during that period.

IV – Conclusions

The evolution of Iberian pig prices is conditioned by internal factors (census, production costs, acorn harvest, etc.) and external factors (the country’s economic situation). In the period of time analysed here (1989-2009), both in the category of piglets and in the category of intensive “Cebo” pigs, 3 price cycles were produced with similar durations; the duration of the first cycle is approximately half of the other two, largely as a result of the programme to eradicate African Swine Fever, and of the country's economic situation.

It is important for price predictions in the Iberian pig market to know the evolution of the country’s GDP, as well as other aspects such as the census and the situations in the farming year.

Acknowledgements

This work has been financed by the [Spanish] Ministry of the Environment & Rural and Marine Affairs, and the Interprofessional Iberian Pig Association (*Asociación Interprofesional del Cerdo Ibérico*). The authors would like to thank the Extremadura Market for their collaboration in completing this work.

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Technology Watch study of the Iberian pig

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Abstract. The Mediterranean Pig is represented in Spain by a group of varieties, strains or ecological adaptations that make up the Iberian pig. We carried out a study on scientific publications in various strategic areas (genetics, nutrition, health, meat and products quality and traceability) related to the Iberian pig. The application of Technology Watch methodologies allows any industry to anticipate market changes and opportunities, understand and reduce the impact of new technologies, overcome access barriers, develop new business ideas, identify potential partners and minimize risk. Several relevant high quality information sources such as scientific publications have been selected using a high-impact database: FSTA (Food Science and Technology Abstract). The information processing required the use of advanced data mining tools (Analyzer Matheo Software). The main findings are reflected in a Technology Sector Map.

Keywords. Technology Watch – Iberian Pig – Technology Map.

Veille technologique pour l'étude du porc Ibérique

Résumé. *Le porc méditerranéen est représenté en Espagne par un ensemble de races et de variations génétiques différentes, issu d'adaptations écologiques, qui composent le porc Ibérique. Nous avons effectué une étude sur des publications scientifiques dans divers domaines stratégiques sur le porc Ibérique (génétique, nutrition, santé, qualité de la viande et des produits ainsi que traçabilité). L'application des méthodologies de veille technologique permet à l'industrie d'anticiper les changements du marché, d'identifier les opportunités, de comprendre et de réduire l'impact lié à l'arrivée de nouvelles technologies, de surmonter les obstacles bloquant l'accès à un projet, de développer de nouvelles pistes liées à l'investissement, d'identifier des partenaires potentiels et de minimiser les risques. De nombreuses données sont sélectionnées à partir de sources d'informations de haute qualité et de pertinence telles que les bases de publications. Nous avons utilisé une base de données spécialisée dénommée FSTA (Food Science and Technology Abstract). Le traitement de l'information nécessite l'utilisation d'outils de data mining avancés (Matheo Analyzer). Une information efficiente est obtenue grâce à l'analyse de cartes retraçant tous les secteurs technologiques du domaine recherché.*

Mots-clés. *Veille Technologique - Porc Ibérique – Carte de la technologie.*

I – Introduction

The current challenge for the jamón *ibérico* (Iberian ham) sector is to provide a product with well defined and constant quality. It is therefore necessary to use the scientific and technological knowledge that has been so far generated and can be applied to serve this remarkable sector in Spain [1].

Technology Watch is the integrated and systematic effort of capturing, analyzing and accurately disseminating and pertinently exploiting all relevant technological and legal information that is necessary for the survival and growth of any institution. In today's knowledge economy, the access to information and its treatment is a prerequisite for the successful undertaking of R&D projects leading to new products, processes or services. The aim of the Technology Watch activities is to alert the company's decision makers of any scientific or technical innovation likely to modify their environment.

II – Methodology

The methodology for this study initiates with the identification of keywords and the narrowing the scope of search. This allows for the retrieval of the relevant information from the selected database. Once a corpus of information is obtained, the analysis and synthesis of the retrieved information is carried out in accordance with the objectives of the study.

The selected database was Food Science and Technology Abstracts, FSTA [2], produced by the International Food Information Service (IFIS). FSTA contains over 500,000 references and covers all areas of food science, food technology, and human nutrition, including basic food science, biotechnology, toxicology, packaging and design. The data mining software Matheo Analyzer [3] was used for the processing of bibliographic records.

III – Analysis of scientific publications

We performed a retrospective study on the research work that has been produced related to Iberian ham. The study covers scientific research published during the period 2000-2010 (August).

1. Scientific indicators

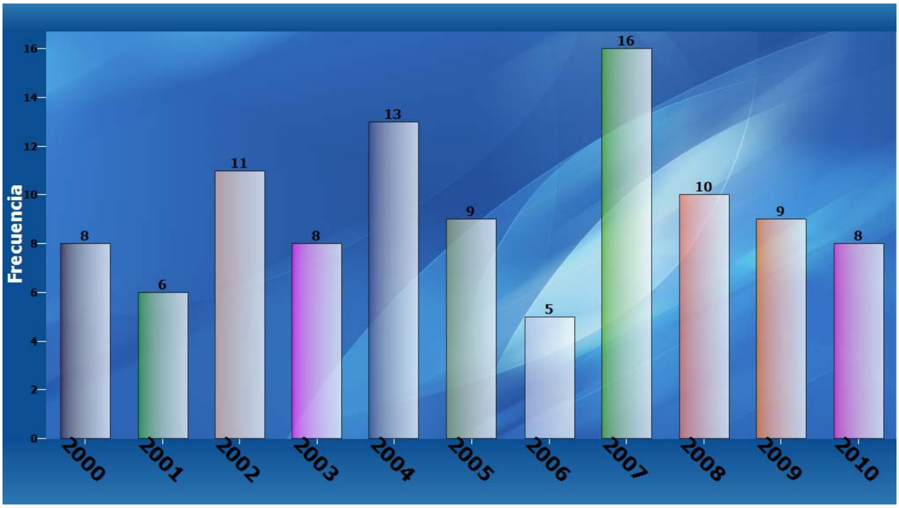

Fig. 1. Evolution of the number of scientific publications (2000-2010).

We obtained 103 scientific publications from FSTA database showing research results related to Iberian ham.

As it can be seen from Fig. 1, the scientific production presents, in general terms, an unstable behavior during the studied period.

Spanish scientific institutions lead the research in the studied area (Table 1); the University of Extremadura holds an absolute leadership, with approximately 59% of all publications retrieved.

Other European institutions that investigate these issues are: Royal Veterinary and Agricultural University and the University of Copenhagen (Denmark), INRA (France) and Fleischerzeugung und fuer Inst Vermarktung (Germany).

Table 2 shows a first group of scientific key terms of the studied area. A total of 251 subject headings describing the different publications research topics were retrieved.

Table 1. Scientific institutions holders

Institutions	TOTAL
Univ. Extremadura	61
Instituto de la Grasa (CSIC)	5
Univ. Complutense of Madrid	4
INIA	3
Univ. of Salamanca	3
IFA-CSIC	2
IMIDA	2
Univ. Zaragoza	2

Table 2. Main scientific content

Subjects	TOTAL
Fatty acids	22
Feeding	21
Ripening	14
Colour	14
Sensory properties	13
Fats animal	13
Genetics	12
Volatile compounds	11
Oxidation	10
Carcasses	9

More recent work on Feeding, for example, focuses on: (i) the Influence of a diet with probiotic bacteria on the lipid composition of Iberian pigs from different tissues; (ii) Individual phospholipid classes from Iberian pig meat as affected by diet (both from the University of Extremadura, 2010).

Table 3 lists topics of new interest related to the study of Iberian ham. Its only appearance in the studied period (2000-2010) is found within the last three years (2008-2010), according to the data contained in FSTA database. The novelty of these terms should be understood in the context of the Iberian ham as the studied subject.

Research on Vacuum, for example, is focused on: (i) modified atmosphere packaging and vacuum packaging for long Period chilled storage of dry-cured Iberian ham; (ii) effect of pressure and holding time on color, protein and lipid oxidation of sliced dry-cured Iberian ham and loin during refrigerated storage; (iii) the effect of HHP treatment (600 MPa) on the oxidative stability of lipids and proteins of vacuum-packaged dry-cured Iberian ham and the Impact on the sensory characteristics of the product was investigated.

Table 3. Emerging scientific content

New topic	2008	2009	2010
Vacuum	0	1	2
Phospholipids	0	0	2
Ultrasound	1	0	0
Triacylglycerols	1	0	0
Swine livers	0	0	1
Subcutaneous fat	0	0	1
Polychlorinated biphenyls	0	0	1
Peptides	0	0	1
Nucleic acids	0	0	1

2. Technology Map

Figure 2 shows a technological map of the Iberian ham for three areas of special interest such as genetics, nutrition and meat quality.

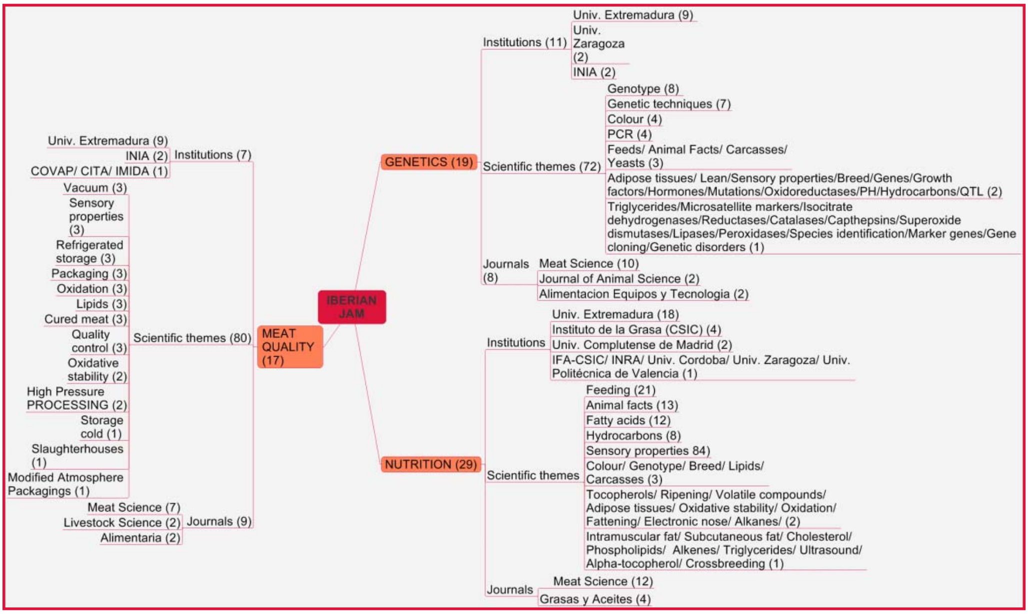

Fig. 2. Technology Map.

IV – Conclusions

The present Technology Watch study has identified key areas of research in the field of Iberian ham from 2000-2010. It enabled to observe the behavior of several selected indicators (evolution of publications, leading institutions, main scientific subjects as well as emerging areas of interest).

The University of Extremadura is the Spanish scientific institution leading research on this area, according to the studied period and the information obtained form the FSTA database.

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[3] Matheo Software, http://www.matheo-software.com/

IBEDROCHES: Socio-economic impact of inter-company cooperation

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Abstract. Ibedroches is a research project involving industrial partners of Iberian hams. Most of the companies are small and medium-sized enterprises (SMEs), located in an area characterized by a mostly agricultural and livestock economy, with a relatively importance of the services sector. In general, their experience in research activities is weak. The main goals of this project are: to characterize the surface mold populations in hams; and to develop and implement the use of spectral sensors in the industry of Iberian pork. A strong community added value will be raised through the culture of cooperation between companies, enhancing socio-economic impacts in the region of El Valle de Los Pedroches. Ibedroches represents an example of how to enhance the confidence of national and international consumers regarding quality of high-value Iberian hams belonging to the region of Los Pedroches, due to its added value as a product manufactured under healthy and sustainable production systems. The development of a tool to ensure product quality objectively will contribute to drive innovation in the fields of promotion and marketing in Iberian ham area. This project is leading an innovation strategy through a consortium of industrial competitors in order to promote added value, employment and economic development in rural areas.

Keywords. Cooperation – Socio-economic impact – Industry – Iberian ham – Rural development.

IBEDROCHES : L'impact socio-économique de la coopération entre entreprises

Résumé. *Ibedroches est un projet de recherche formé par différentes entreprises de jambon Ibérique. La plupart d'entre elles sont des petites et moyennes entreprises (PYMES). Elles se trouvent dans une zone dont l'économie est principalement basée sur l'agriculture et l'élevage, avec une importance relative du secteur des services. En général, l'expérience patronale en matière d'activités de recherche est faible. Les objectifs principaux du projet sont: caractériser les populations superficielles de moisissures du jambon; développer et implanter l'usage de capteurs spectraux dans l'industrie du porc Ibérique et obtenir finalement des résultats qui fortifient les valeurs socio-économiques dans la vallée de Los Pedroches, grâce à la coopération entre les entreprises du secteur. Ibedroches représente un exemple montrant comment augmenter la confiance des consommateurs nationaux et internationaux concernant la qualité de pièces de jambon exquises, qui ont une haute valeur, produites dans la vallée de Los Pedroches, surtout en raison de leurs valeurs ajoutées de produit sain, provenant de systèmes durables de production. Le développement d'un outil permettant de garantir une qualité plus objective du produit, aidera à l'innovation pour les systèmes de promotion et de commercialisation du jambon Ibérique. Il s'agit d'un projet où l'innovation principale consiste à créer une valeur ajoutée, c'est-à-dire à favoriser l'emploi et le développement socio-économique en zones rurales, au moyen de la R+D+I promue par l'association entre des entreprises qui sont, en principe, concurrentes.*

Mots-cles. *Coopération – Facteurs socio-économiques – Jambon Ibérique – Développement.*

I – Introduction

The livestock population across the EU-27 comprised about 161 million pigs, 95 million sheep, and 88 million head of cattle in 2006, as well as 1.5 billion poultry birds (Eurostat, 2008). Pig production is specialized even across borders, with breeders such as Denmark, fatteners such as Spain and mixed producers such as the Netherlands. Germany, Denmark and the Netherlands form a single pig production area (Marquer, 2010). In 2008 pork production in the EU-27 reached 259.6 million head, of which more than half (54.4%) came from four countries

(Germany, Denmark, Spain and France). Otherwise, Eurostat report (2008) shows that the highest annual apparent consumption among meat products was recorded for pork products, averaging over 40 kg per capita, a level that was higher than the combined total of poultry, cattle, sheep and goats. Spain, Austria, Germany, Denmark and Belgium reported the highest per capita apparent consumption of pig meat, all recording averages in excess of 50 kg.

Pork is produced throughout the EU on several types of farms with considerable variations from one Member State to another. In southern European countries most of the dry-cured meat products come from conventional or improved pig breeds reared indoors and fed on commercial feed mixtures. In some areas of the Mediterranean basin, pig production relies on local breeds (Iberian, Corsican) whose expend a final extensive fattening period in the Mediterranean forest. On the other hand, there is an increasing demand for both fresh and dry-cured products from Iberian pigs reared under intensive nutritional management, due to their high quality at a comparatively lower price (García-Valverde *et al*., 2008).

The meat industry sector is characterized by a elevated number of small and medium enterprises (SMEs), in particular Iberian hams industries, most of them with a reduced number of employees. The sector is highly atomized (Cruz, 2009). Because of that, together to their strong traditional character, it is difficult to find meat companies that employ part of their resources to R&D activities.

Previous reports strength the interest on a competitive improvement of the meat industries faced to a strongly competitive market and the panorama of economic crisis (UTEDLT, 2008; www.infocarne.com). Therefore, the promotion of the innovation in the meat industry is a sectorial strategic activity. The development of managerial projects of investment in R&D, individuals or in cooperation, is fundamental in order to assure the survival and long-term viability of the industries.

Ibedroches is a research project involving six industrial partners of Iberian hams in the region of Los Pedroches (Córdoba, Spain). The main goals of this project are to obtain a strong community added value through the culture of cooperation between companies, enhancing socio-economic impacts, as well as through improving the quality and food safety of the Iberian ham.

II – Materials and methods

The consortium involves six meat companies: Camilo Ríos, Celestino Gómez, COVAP, Hnos. Rodríguez Barbancho, IBESA, La Finojosa, under the coordination of CICAP and in collaboration with the Universities of Córdoba and Extremadura.

Five of the six companies are SMEs, located in an area characterized by a mostly agricultural and livestock economy, with a relatively importance of service sector. All of them produce Iberian meat products from pigs reared in the Mediterranean forest. In general, their experience in research activities is weak.

The six companies, with industrial activity in the same geographic area (region of Los Pedroches), are working as a group in two different activities packages: toxigenic molds control and evaluation of spectral technologies EIS and NIRS.

III – Results and conclusions

Ibedroches represents an example of how to enhance the confidence of national and international consumers regarding quality of high-value Iberian hams belonging to the region of Los Pedroches, due to its added value as a product manufactured under healthy and sustainable production systems. The development of a tool to ensure product quality objectively will contribute to drive innovation in the fields of promotion and marketing in Iberian ham area. This project is leading an innovation strategy through a consortium of industrial competitors in order to promote added value, employment and economic development in rural areas.

The added value obtained from the present study will enhance the quality and food safety of the Iberian ham reducing production costs, becoming in a useful tool for the meat industries. Thus, the technological development derived from the close work of the companies' participants in the project will generate a portable tool that might differ these companies from the competition. This device will be able to be patented by the corresponding economic benefit. All these results expect to open or consolidate new frontiers of the more demanding countries, such as United States or Japan.

Acknowledgments

The authors would like to thanks to: UCO, UNEX, Embutidos Camilo Ríos, Celestino Gómez, COVAP, Hnos. Rodríguez Barbancho, IBESA, Jamones y Embutidos La Finojosa. The project is financed by European regional development fund (ERDF) in the form of "Fondo Tecnológico" Ref. IDI 20091171 to IDI 20091176.

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OPTIONS méditerranéennes

SERIES A: Mediterranean Seminars
2012 – Number 101

7th International Symposium on the Mediterranean Pig

Edited by:
E.J. De Pedro and A.B. Cabezas

This publication presents the Proceedings of the 7th International Symposium on the Mediterranean Pig held in Cordoba (Spain) from 14 to 16 October 2010. More than twenty years have passed since the first symposium was organized in 1989 in Ajaccio (Corsica), with the participation of French, Italian, Portuguese and Spanish researchers. Since then, several symposia have been held and researchers from other European regions and from the rest of the world have joined the group, aiming to share, discuss and spread the scientific progress achieved in pig production linked to sylvopastoral and agroforestry systems.

The objective of the Network on the Mediterranean Pig is to contribute to the progress of this production system, which brings to society all the values it possesses: it is a key element in the economic development of rural areas, it plays a key role in the environmental management of zones associated with sylvopastoral and agroforestry systems, and it offers high-quality products, demanded by consumers and very much desired by other types of livestock production systems.

The pig production system must be studied within a wider context and, thus, issues related to genetics, production management, nutrition, health, product and socioeconomic aspects have been dealt with in the Symposium. This volume contains a total of 114 articles from contributions presented at the Symposium on all those topics. Given that this product quality plays an important role in the survival of these systems, 40% of the work has dealt with this aspect, as well as the traceability of the systems (12%), with the aim of contributing transparency to commercial transactions and giving a guarantee to the consumers in the different stages of the system (stockbreeders, industry, consumers).

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CIHEAM
MAI ZARAGOZA

ISBN: 2-85352-488-4
ISSN: 1016-121-X

Prix: 106,71 Euro

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