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in
5. International Symposium on the Mediterranean Pig
Zaragoza : CIHEAM
Options Méditerranéennes : Série A. Séminaires Méditerranéens; n. 76
2007
pages 155-158

Article available on line / Article disponible en ligne à l’adresse :
http://om.ciheam.org/article.php?IDPDF=800576

To cite this article / Pour citer cet article
α-tocopherol content and susceptibility to lipid peroxidation in muscle from Iberian pigs fed in two different "recebo" systems

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SUMMARY – The α-tocopherol content and susceptibility to lipid peroxidation in Gluteo biceps muscle from Iberian $x$ Duroc (50%) pigs were analysed. The α-tocopherol content was higher (p<0.01) in muscle from recebo 2 pigs, which consumed higher quantities of vitamin E enriched concentrate feed (220 mg/kg) during the fattening period, than in recebo 1 pigs. Susceptibility of the muscle to lipid oxidation was significantly (p<0.01) influenced by the feeding system, animals reared in recebo 1 system showing higher levels of MDA after 50 and 100 min of incubation than recebo 2 pigs. These results indicate that α-tocopherol content in the muscle depends on the levels of this natural antioxidant provided by dietary means.

Keywords: Iberian pig, meat, tocopherols, peroxidation.

RESUME – "Teneur en α-tocophérol et susceptibilité à la péroxydation lipidique de muscles de porcs Ibériques alimentés selon deux systèmes différents de «recebo»". Dans ce travail, nous avons analysé la teneur en α-tocophérol et la susceptibilité à l’oxydation des lipides du muscle Gluteo biceps de porcs Ibériques $x$ Duroc (50%). La quantité de α-tocophérol dans le muscle était supérieure (p<0,01) dans les muscles des porcs du lot recebo 2, qui ont consommé des quantités supérieures de concentrés enrichis en α-tocophérol/acetate (220 mg/kg) pendant la période d’engraissement. La susceptibilité du muscle à l’oxydation était influencée significativement (p<0,01) par le système d’alimentation. Les porcs alimentés avec le système recebo 1 ont présenté des niveaux de MDA après 50 et 100 minutes d’incubation supérieurs à ceux des porcs du lot recebo 2. Ces résultats montrent que la quantité de α-tocophérol dans le muscle dépend des niveaux de cet antioxydant naturel apportés par l’alimentation.

Mots-clés : Porc Ibérique, viande, tocophérols, péroxydation.

Introduction

Feeding during the last period of life prior to slaughter is the main factor determining the high acceptability of meat and meat products from Iberian pig (García et al., 1996; Cava et al., 2000a). The highest quality of dry-cured products is obtained using meat from pigs reared outdoors (usually known as montanera system, with feeding based on acorns and pasture), whereas the lowest quality is obtained when using pigs reared indoors (feeding based on concentrate feeds). Within each commercial category there are some factors that influence Iberian pig products characteristics, such as the outdoors fattening period length (Flores et al., 1988) or the concentrate feeding composition (Cava, et al., 1999).

At present, management system known as recebo, in which feeding during the fattening period is based on acorn and pasture and by feeding with concentrate feeds, is one the most practiced production system in Iberian pig sector. This system allows to increase the number of produced pigs, enabling the obtention of high quality products. Products from Iberian recebo pigs are commonly considered as intermediate quality products (between products from pigs reared outdoors and from pigs reared indoors) (García et al., 1996).

In recent years, two strategies are being carried out in diet formulation for Iberian pigs fattening, taking into account the quality characteristics demanded for the final product. These strategies are based on the use of fat-enriched diets (up to 7-9%), based principally on feeds enriched with oleic acid (C18:1), allowing a similar fatty acid composition to that of acorns, and on the use of α-tocopherol enriched diets in order to increase the level of this antioxidant in muscles, which is an effective mean for reducing lipid oxidation in fresh meat and in meat products (Monahan et al., 1992a; Cava et al., 1999; Isabel et al., 2003).
The aim of this work was to evaluate the content of α-tocopherol and the susceptibility to lipid oxidation of meat from Iberian pigs reared in two different production systems, based on acorn and pasture and completed the fattening period with concentrated feed, systems known as recebo.

**Materials and methods**

**Animals and diets**

Iberian x Duroc (50%) pigs (n=26) were fed on two different recebo systems (recebo 1 and recebo 2). Differences between the two systems were based on the amount of acorn and pasture consumed by pigs and content on concentrate feeding (enriched with oleic acid and α-tocopherol) supplemented. The pigs from recebo 1 group were raised outdoors during 67 days consuming mainly acorn (60% of acorn from *Quercus suber* and 40% from *Q. rotundifolia*), pasture (management system named as montanera) and 0.7 kg of concentrate feed per day. Finally, to conclude fattening process pigs were fed exclusively with 3.5 kg/day of concentrate feed during 20 days. Pigs from recebo 2 group were raised outdoors during 55 days consuming acorn, pasture and 1 kg/day of concentrate feed, concluding the fattening period with 3.7 kg/day of concentrate feed during 50 days. The concentrate feed consisted basically on cereals and was enriched with olein (6%) and α-tocopheryl-acetate (220 mg/kg). Pigs were slaughtered at 170±8 kg live weight at a local slaughterhouse.

**Sampling**

Sampling was carried out within 24 h after slaughter. A portion of the *Gluteo biceps* muscle from raw hams were taken for analysis. Muscle samples were trimmed of visible fat and immediately placed under vacuum in a freezer and stored at -80°C until analysis.

**Determination of α-tocopherol in Muscle**

α-tocopherol in muscle were determined following the method described by Rey *et al.* (1996). 0.8 g muscle were homogenized in 6 ml 0.054 M dibasic sodium phosphate buffer adjusted to pH 7.0 with HCl. After mixing with absolute ethanol and hexane, the upper layer containing tocopherol was evaporated to dryness and solved in 200 µl ethanol prior to analysis by reverse phase HPLC (Agilent 1100 Series, with a Diode Array detector). Separation was made on an Agilent Technologies Lichrospher RP-C18 column (250 mm x 4 mm i.d., 5 µm particle size), the mobile phase was methanol:water (97:3 vol/vol) at a flow rate of 2 ml/min and peaks were registered at 292 nm. Peaks were identified and quantified upon calibration with standards of α-tocopherols (Sigma Chemical Co., St. Louis, USA).

**Measurement of induced lipid oxidation**

The susceptibility of muscle tissue homogenates to iron-ascorbate-induced lipid oxidation was determined by a modification of the method of Kornbrust and Mavis (1980), in which 1 mM FeSO₄ was used as the catalyst of lipid oxidation. Homogenates (approximately 50 mg/ml buffer) were incubated at 37°C in 40 mM tris-malate buffer (pH 7.4) with 1 mM FeSO₄ in a total volume of 10 ml. At fixed time intervals, 1 ml aliquots were removed for measurement of thiobarbituric acid-reactive substances (TBARS). TBARS were expressed as nM malondialdehyde (MDA)/mg protein. Protein was measured by the procedure of Lowry *et al.* (1951).

**Statistical analysis**

The effect of rearing conditions on α-tocopherol concentration and lipid induced oxidation was analysed using a General Linear Model of SPSS v.11.5 (2003).
Results and discussion

The α-tocopherol content (Fig. 1) of Gluteo biceps muscle from Iberian pigs fed on extensive system together with concentrated diets supplemented with 220 mg/kg α-tocopheryl acetate (recebo 1 and recebo 2) varied from 4.67 µg to 5.91 µg. The α-tocopherol content was higher (p<0.01) in muscle from pigs that consumed higher quantities of vitamin E enriched concentrate feed (220 ppm) during the fattening period. These values are higher than other previously reported in literature by different authors in Iberian pigs fed outdoors with acorn and pasture (Cava et al., 2000b) or exclusively with α-tocopheryl acetate supplemented diets (Isabel et al., 1999; Cava et al., 2000b). Several authors have evidenced the positive relationship between consumption and deposition of α-tocopherol in muscle of Iberian pigs (Cava et al., 2000b; Isabel et al., 1999) and in other pig breeds (Jensen et al., 1997; Monahan et al., 1992).

The effect of the two recebo systems used in this study on susceptibility of Gluteo biceps lipid fraction to oxidation assessed by induced peroxidation is also shown in Fig. 2. Susceptibility to lipid peroxidation was also significantly (p<0.01) affected by recebo system performed in this study. Muscles homogenates from recebo 1 pigs show higher susceptibility to lipid peroxidation than muscles homogenates from recebo 2 pigs. Therefore, it can be concluded that differences in the susceptibility of muscle homogenates to lipid oxidation are clearly related to muscle levels of α-tocopherol, in accordance with studies carried out previously (Isabel et al., 2003; Hoz et al., 2003).

Conclusions

In conclusion, dietary α-tocopherol supplementation during the fattening period of Iberian pigs produced in recebo system increased the antioxidant status of Iberian pig muscles.
References


Fig. 2. Susceptibility to lipid peroxidation in Gluteo biceps muscle from Iberian x Duroc pig.