

Fatty acids analysis of Iberian pig fat by near infrared spectroscopy (NIRS)

García-Olmo J., Pedro E. de, Garrido-Varo A., Jiménez A., Salas J., Santolalla M.

in

Almeida J.A. (ed.), Tirapicos Nunes J. (ed.).
Tradition and innovation in Mediterranean pig production

Zaragoza : CIHEAM

Options Méditerranéennes : Série A. Séminaires Méditerranéens; n. 41

2000

pages 191-195

Article available on line / Article disponible en ligne à l'adresse :

<http://om.ciheam.org/article.php?IDPDF=600133>

To cite this article / Pour citer cet article

García-Olmo J., Pedro E. de, Garrido-Varo A., Jiménez A., Salas J., Santolalla M. **Fatty acids analysis of Iberian pig fat by near infrared spectroscopy (NIRS)**. In : Almeida J.A. (ed.), Tirapicos Nunes J. (ed.). *Tradition and innovation in Mediterranean pig production*. Zaragoza : CIHEAM, 2000. p. 191-195 (Options Méditerranéennes : Série A. Séminaires Méditerranéens; n. 41)



<http://www.ciheam.org/>
<http://om.ciheam.org/>

Fatty acids analysis of Iberian pig fat by near infrared spectroscopy (NIRS)

J. García-Olmo*, E. De Pedro*, A. Garrido-Varo*, A. Jimenez**, J. Salas** and M. Santolalla**

*Departamento de Producción Animal, ETSIAM, Universidad de Córdoba,
P.O. Box 3.048, 14080 Córdoba, Spain

**Laboratorio Agroalimentario, Consejería de Agricultura y Pesca de Andalucía,
Avda. Menéndez Pidal s/n, 14080 Córdoba, Spain

SUMMARY - The objective of the present work was to obtain NIR calibration equations for the prediction of the content of fatty acids in subcutaneous fat from Iberian pigs. Calibration equations were obtained, using 352 samples of fat from animals belonging to Designation of Origin "Jamón de Huelva". The precision of the equations was evaluated by the coefficient of determination (R^2) and standard errors of cross validation (SECV). Both were excellent for the fatty acids studied, with R^2 of 0.96, 0.97, 0.99 and 0.91, and SECV (%) of 0.28, 0.24, 0.25 and 0.18, for palmitic, stearic, oleic and linoleic acid, respectively. The precision of the equations was then confirmed by external validation on a set of 20 samples.

Key words: Iberian pig, fatty acids, NIRS, folded transmission.

RESUME - "Teneurs en acides gras du porc Ibérique obtenues par NIRS". Ce travail a pour objectif l'obtention d'équations de calibration NIRS pour la prédiction des teneurs en acides gras du tissu adipeux des carcasses du porc Ibérique. L'étude a été réalisée sur 352 échantillons de gras d'animaux dont l'Appellation d'Origine est "Jamon de Huelva" et qui ont été abattus durant les campagnes 96/97 et 97/98. La précision des équations obtenues selon les paramètres statistiques R^2 : (Coefficient de détermination) et ETVC (Erreur type de validation croisée) s'avère excellente pour les quatre acides gras étudiés. En effet, ces derniers sont comme suit : $R^2 = 0,96$ et ETVC = 0,28% pour l'acide palmitique, $R^2 = 0,97$ et ETVC = 0,24% pour l'acide stéarique, $R^2 = 0,99$ et ETVC = 0,25% pour l'acide oléique et $R^2 = 0,91$ et ETVC = 0,18% pour l'acide linoléique. La haute précision des équations obtenues a été confirmée lorsque ces dernières s'appliquent sur un collectif de validation externe constitué par 20 échantillons.

Mots-clés : Porc Ibérique, acides gras, NIRS, folded transmission.

Introduction

Several Designations of Origin (DO) of Iberian ham, "Guijuelo", "Dehesa de Extremadura", "Jamón de Huelva" and "Pedroches", private processing industries and the Spanish pig producers associations have established quality control programmes to determine the type of feeding used for Iberian pigs, especially during the final fattening period (montanera). These programmes include on-farm inspection, for example of the weight and number of animals, length of the montanera, quantity of acorns eaten, and analysis of the contents of the fatty acids in subcutaneous fat.

At the same time, at the request of the Interprofessional Association for the Iberian Pig (ASICI), the Ministry of Agriculture and Fisheries established the so-called "homologated contract to fix the price for slaughtering Iberian pigs". This is published every year in the National Official Journal (BOE) and establishes quality specifications based on weight, type of feeding and content of the palmitic, stearic, oleic and linoleic acids in the subcutaneous fat. The quality specifications allow the classification of the animals into three commercial categories, "bellota", "recebo" and "pienso", with different prices.

The high cost of farm recording and of analysis of subcutaneous fat by gas chromatography restricts the extension of these quality control programmes to all of the animals produced. As a consequence, only a representative sample of a group of animals is analysed, sampling oscillates between 20% and 50% of the pigs in every group, depending on the DO or company.

Basic research by the Department of Animal Production of the University of Cordoba has shown the possibilities of Near Infrared Spectroscopy (NIRS) as a technique for analysing and

characterizing Iberian pig fat in relation to the type of feeding (De Pedro *et al.*, 1992, 1995, 1997; Hervás *et al.*, 1994). The main advantages of this technique are that an instantaneous analysis is possible, it is non destructive, non contaminating and allows several constituents to be predicted simultaneously (Garrido *et al.*, 1996).

The objective of the present work was to obtain calibration equations to predict the content of the fatty acids in the subcutaneous fat of Iberian pigs produced under the DO "Jamón de Huelva".

Materials and methods

Samples

A total of 352 samples of subcutaneous fat of Iberian pigs produced under the DO "Jamón de Huelva" regulations during 1997 and 1998 were used. Liquid fat samples were analysed, using Gas Chromatography and Near Infrared Spectroscopy after fusion of adipose tissue samples by microwaves (De Pedro *et al.*, 1996).

Analysis of fatty acids by Gas Chromatography

The determination of the content of the fatty acids by gas chromatography of representative liquid fat samples was made by the Laboratorio Agroalimentario of Junta de Andalucía, in Córdoba. The methyl esters of fatty acids were extracted with hexane and were determined, using a Perkin-Elmer Sigma 3D with FID detector.

NIRS analysis

Spectra were obtained by folded transmission, from 400 to 2500 nm, with a cell for analysis of liquid products of 0.1 mm pathlength (ref. IH- 0345), using a spectrophotometer Foss-NIRSystem 6500 with a spinning module. Spectra were collected, using the software ISI NIRS 3 version 3.11 (Infrasoft International, Port Matilda, PA, USA).

Chemometric analysis of NIRS data

The ISI NIR 3 software ver. 3.11. was used for the chemometric analysis of NIRS data, with methodology for development and validation of NIRS calibrations described by Mark and Workman (1991), Shenk and Westerhaus (1995 and 1996) and Williams and Sobering (1996).

Statistics used to select the best calibration equations were: standard error of calibration (SEC), standard error of cross validation (SECV) and standard error of prediction (SEP); the standard error of the reference method (SEL) or standard error of duplicates analysed by the reference method (gas chromatography); coefficient of determination of calibration (R^2) and cross validation (r^2); RPD or relation between the standard deviation of the reference data and the standard error of cross validation and RER or relation between the range of the reference data and the standard error of cross validation.

Results and discussion

Table 1 shows mean contents of the fatty acids in calibration samples. Chemometric treatment of data allows different NIRS calibration equations for each fatty acid to be developed. NIRS equations were selected, taking into account the best statistical values of r^2 and SECV (Garrido *et al.*, 1996; Shenk and Westerhaus, 1996,) and other statistics (RPD and RER), of interest from the point of view of the use of a particular equation at the industry level (Williams and Sobering, 1996). SEL was obtained by duplicated analysis of 20 samples, using the reference method.

Table 1. Fatty acids composition of calibration set

	n	Mean	Sd	Range	SEL
%C16:0	352	21.1	1.5	17.4-25.3	0.26
%C18:0	352	10.7	1.3	7.7-14.9	0.22
%C18:1	352	52.3	2.4	45.0-58.1	0.25
%C18:2	352	9.4	1.3	6.8-13.5	0.15

Sd: standard deviation; SEL: standard error of reference method

Table 2 summarizes statistical values for the best calibration equations obtained. Higher values of the coefficient of determination of calibration (R^2) and cross validation (r^2) were obtained for the four fatty acids studied. Shenk and Westerhaus (1996) indicate that NIRS equations with coefficient of determination values higher than 0.9 may have an excellent precision and those between 0.5 and 0.9 have values of a good precision.

Table 2. Statistics of calibration equations (n=352)

	Mean	Sd	SEC	R^2	SEVC	r^2	RER	RPD
%C16:0	21.09	1.46	0.24	0.97	0.28	0.96	28.2	5.2
%C18:0	10.68	1.33	0.19	0.98	0.24	0.97	30	5.5
%C18:1	52.31	2.44	0.22	0.99	0.25	0.99	52.4	9.8
%C18:2	9.39	1.32	0.15	0.99	0.18	0.98	37.2	7.3

Sd: standard deviation; SEC: standard error of calibration; SECV: standard error of cross validation; R^2 : coefficient of determination of calibration; r^2 : coefficient of determination of cross validation

Table 2 shows that all the coefficients of determination values of cross validation are excellent, reaching values close to 1 in the case of the main fatty acid (oleic acid). The SECVs, are similar to the standard errors (SEL) of the reference method.

RPD and RER statistics confirm the high precision of the equations developed, with values higher than the minimum values recommended by Williams and Sobering (1996) for both statistics (RPD>3 and RER>10).

Once the equations with the best statistics were obtained, they were applied to an external validation set of 20 samples, from animals reared during 1998 and not included in the calibration set. As can be seen in Table 3, the fatty acid composition of samples of the external validation set, is similar to the composition of samples of the calibration set (Table 2).

Table 3. Fatty acids composition of external validation set

	n	Mean	Sd	Range
%C16:0	20	20.1	1.6	17.4-23.4
%C18:0	20	9.5	1.3	7.6-12.4
%C18:1	20	53.6	2.7	47.4-58.1
%C18:2	20	10.1	1.2	8.6-12.4

Sd: standard deviation

Figure 1 shows NIRS predicted data versus reference method (GC) data, with standard error of prediction (SEP) and coefficient of determination of the external validation set (R^2). These results confirm the excellent correlation between the methods.

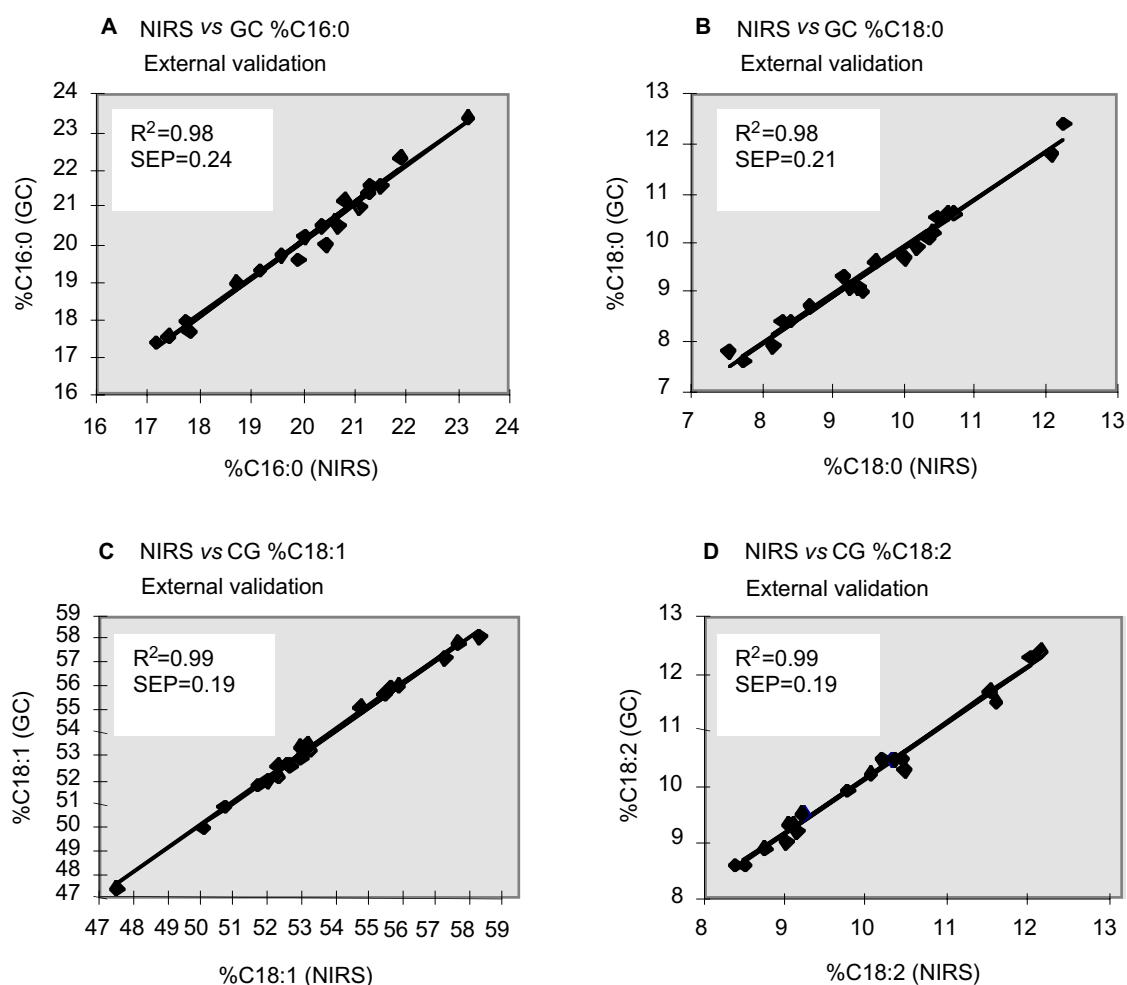


Fig. 1. NIRS predicted values *versus* GC values.

Conclusions

(i) NIRS equations have a high precision, when evaluated by the coefficient of determination (r^2), the standard error of cross validation (SECV), RER and RPD.

(ii) The application of NIRS technology to industries and laboratories, would allow quality control to be extended to all the animals of one group, and at less cost than current quality control, using gas chromatography.

Acknowledgements

The authors wish to thank DO "Jamón de Huelva", for providing the fat samples. All NIRS data were obtained, using the instrumental and software of the NIRS Centralized Unit of the University of Córdoba. This work was supported by the Junta of Andalucía.

References

- De Pedro, E., Casillas, M. and Miranda, C.M. (1996). Microwave oven application in the extraction of fat from the subcutaneous tissue of Iberian pig ham. *Meat Sci.*, 45(1): 45-51.
- De Pedro, E.J., Garrido, A., Bares, I., Casillas, M. and Murray, I. (1992). Application of near infrared spectroscopy for quality control of Iberian pork industry. In: *Near Infrared Spectroscopy. Bridging the Gap between Data Analysis and NIR Applications*, Hildrum, K.I., Isakson, T., Naes, T. and Tandberg, A. (eds). Ellis Horwood, NY, pp. 341-347.
- De Pedro, E., Garrido, A., Lobo, A., Dardenne, P. and Murray, I. (1995). Objective classification of Iberian pig carcasses: GC versus NIR. In: *Leaping ahead with Near Infrared Technology*, Batten, G.D., Flin, P.C., Welsh, L.A. and Blankeney, A.B. (eds). Royal Australian Chemical Institute, Victoria, Australia, pp. 291-295.
- De Pedro, E., Garrido, A., Martínez, M.L., Angulo, F. and García-Olmo, J. (1997). Espectroscopia de Infrarrojo Cercano en el análisis cuantitativo y cualitativo de productos derivados del cerdo Ibérico. *ITEA*, 18: 661-663.
- Garrido, A., Gómez, A., Guerrero, J.E. and Fernández, V. (1996). NIRS: Una tecnología de apoyo para un servicio integral en Alimentación Animal. In: *Avances en Alimentación y Nutrición Animal*, García, P., González, G. and De Blas, C. (eds). FEDNA, pp. 275-300.
- Hervás, C., Garrido, A., Lucena, B., García, N. and De Pedro, E. (1994). Near infrared spectroscopy of Iberian pig carcasses using an artificial neural network. *Journal of Near Infrared Spectroscopy*, 2: 177-184.
- Mark, H. and Workman, I. (1991). *Statistics in Spectroscopy*. Academic Press, Inc., NY.
- Shenk, J.S. and Westerhaus, M.O. (1995). *Routine operation, calibration, development and network system management manual*. NIRSystems, Inc., 12101 Tech Road, Silver Spring, MD 20904, PN IS-0119.
- Shenk, J.S. and Westerhaus, M.O. (1996) Calibration the ISI way. In: *Near Infrared Spectroscopy: The Future Waves*, Davies, A.M.C. and Williams, P.C. (eds). NIR Publications, Chichester, UK, pp. 198-202.
- Williams, P.C. and Sobering, D. (1996). How do we do it: a brief summary of the methods we use in developing near infrared calibrations. In: *Near Infrared Spectroscopy: The Future Waves*, Davies, A.M.C. and Williams, P.C. (eds). NIR Publications, Chichester, UK, pp. 185-188.