CHANGES IN LIPID AND NITROGEN FRACTIONS IN DRY-CURED PORK FORELEG ("LACÓN GALLEGO") AFTER DESALTING AND COOKING.

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Background

Dry cured pork foreleg "Lacón gallego" is a typical product manufactured in the northwest of Spain where it has a great economical importance. In 2001, Protected Geographical Indication (PGI) "Lacón Gallego" was recognised in European Union (Commission of the European Communities, 2001). Its elaboration is begun cutting the fore extremity of the pig at the shoulder blade-humerus joint. Processing involves a series of phases: salting, washing, standing and drying or curing in a minimum of thirty days (approximately 35 days). The final product is normally eaten after desalting and cooking (boiling).

Proteolysis and lipolysis are important phenomena occurring during dry cured meat products manufacture; it influences final sensory properties due to the generation of low molecular weight compounds which generate the flavour or are flavour precursors (Toldrá, 1998). Protein breakdown involves increases in non protein nitrogen concentration in dry cured hams and in smaller fractions, such as free amino acids (Buscaihlon *et al.,* 1994; Martín *et al.,* 1998). An intense lipolysis has been observed especially in the first months of dry cured ham processing; free fatty acids produced are precursors of volatile compounds responsible of flavour (Toldrá, 1998).

Cooking produces important changes in meat compounds. The formation of volatile flavour compounds due to Maillard reactions, in which free amino acids are involved, generally occur at temperatures associated with cooking (Whitfield, 1992). Lipid oxidation is also influenced by cooking (Morrisey *et al.,* 1998) and fat-soluble carbonyls contribute to meat flavour (Sanderson *et al.,* 1966). On the other hand, proteolytic and lipolytic changes could occur during cooking; some endogenous enzymes, such as cathepsins B and L, can generate flavoured small peptides (Spanier *et al.,* 1988) during heating and show activity in late stages of dry cured ham processing (Toldrá, 1998). Against this background, it is possible that changes in lipid and nitrogen fractions occur during cooking of "Lacón Gallego"; however, there are no studies about this topic.

Objectives

The objective of this work was to study the effect of desalting and cooking on lipid and nitrogen fractions of dry-cured foreleg protected by PGI.

Methods

Ten crossbred pigs were fattened and slaughtered controlled by PGI Council inspectors. Ten fore extremities were removed from these carcasses post-slaughter at the shoulder blade-humerus joint. The raw pieces (about 4 kg. each one) were rubbed with salt, containing about 0.1% potassium nitrate, and placed in piles of salt at low temperature (2-5ºC) and high relative humidity (80-90%) for 5 days. After washing to remove salt from the surface, the pieces were hung in a post-salting room at 2-5ºC and relative humidity of 85% for 15 days. Once the post-salting stage had finished, the pieces were transferred to a drying-ripening room at 12ºC and 60% of relative humidity during 15 days. Five dry-cured forelegs were removed for testing at the end of the processing. The samples were taken from 2 different zones of the foreleg, representing external or superficial muscles (*m. pectoralis profundus, m. serratus ventralis, m. supraspinatus* and *m. subscapularis*) and internal or deep muscles (*caput lateral mi. tricipitis brachii, caput longum mi. tricipitis brachii, m. infraspinatus and m. deltoideus*). Samples were vacuum packaged and kept at -20ºC until analysed.

The other five dry-cured forelegs were desalted in water during 48 hours. Afterwards, the desalted forelegs were cooked in boiling water during 60 minutes. The samples were also taken from 2 different zones of the foreleg, representing external and internal muscles as described in dry-cured forelegs.

Samples were finely minced in a blender (Polytron PT 10-35). Lipids were extracted and purified from the former homogenate according to the method described by Hanson and Olley (1963). Total intramuscular lipids were fractionated into neutral lipids, free fatty acids and phospholipids on NH_2-aminopropyl minicolumns according to the method described by García-Regueiro et al. (1994). The neutral lipid fraction contained mostly glycerides and this term will be used throughout the text. Amounts of glycerides, phospholipids and free fatty acids were expressed as percent of total fat weight obtained.

For the determination of the various nitrogen fractions, a portion of minced dry cured or cooked foreleg (10g) were homogenized with 50 ml of 0.6N perchloric acid and centrifuged (1700 x g for 10 min, 4ºC). The pellet was reextracted in the same conditions. The volume of combined supernatants was adjusted to 100 ml. Non protein nitrogen (NPN) was determined following the Johnson (1940) method. Phosphotungstic acid soluble (PTN) and sulphosalicylic acid soluble (SSN) nitrogens were obtained from the 0.6N perchloric acid soluble fraction as previously described (Díaz *et al.,* 1992). Samples for ammonia analysis were prepared according to Barchietto *et al.* (1984). Ammonia levels were determined using the Sigma kit for enzyme analysis (Sigma-Aldrich Chemical) following the manufacturer instructions.

Statistical treatment was performed by using Student´s t-test for comparison between means of dry-cured foreleg and cooked dry-cured foreleg for external and internal muscles (SPSS version 9.0.1. for Windows, 1998).

Results and discussion

The lipid fraction composition of meat from forelegs is shown in Table 1. The glycerides of dry-cured forelegs represented 74-75%, the phospholipids 13-14% and the free fatty acids 11-12% of the total lipid content, respectively. The contents of free fatty acids in cooked dry-cured forelegs were 18% in external muscles and 16% in internal muscles. So, the desalting and cooking of dry-cured forelegs increased the free fatty acid contents. The contents of glycerides and phospholipids were lower in cooked dry-cured forelegs (71% and 11% respectively) than in dry-cured forelegs. These results reveal that there are hydrolysis in glycerides and phopholipids.

Table 2 shows the changes in nitrogen fractions due to cooking process. Nitrogen fractions contents in dry cured forelegs are in the range of values reported by Marra *et al.* (1999), although these authors did not analysed internal and external muscles separately. NPN, PTN and SSN decreased in cooked dry cured samples, both external and internal muscles, although significant differences were only found in PTN and SSN in internal muscles. The highest decreases were observed in PTN and SSN, which could be attributed to chemical reactions with other compounds mainly in internal muscles. These reactions (e.g. Maillard reactions) have been observed in cooked meats (Whitfield, 1992). In external muscles samples it could be also attributed to losses in the water during boiling. Ammonia increased

in cooked samples, although it was only significant in internal muscles. It could probably due to amino acids deamination during cooking process (Flath *et al.,* 1981).

Conclusions

Cooking of dry cured pork forelegs increased significantly free fatty acids in all samples and ammonia content in internal muscles, while the other nitrogen fractions decreased.

Pertinent literature

BARCHIETTO, G., CANTONI, C., FRIGERIO, R. & PROVERA, D. 1984. Esame comparativo del prodotti di autolisi nella carne di maiale (azoto non proteico, urea, ammoniaca). Conservazienoni degli Alimenti 3, 12-17.

BUSCAILHON, S., MONIN, G., CORNET, M. & BOUSSET, J. (1994). Time related changes in nitrogen fractions and free amino acids of lean tissue of French dry-cured ham. Meat Sci. 37, 449-456.

COMMISSION OF THE EUROPEAN COMMUNITIES (2001). Commission Regulation (EC) Nº 898/2001 of 7 May supplementing the Annex to Regulation (EC) Nº 2400/96. Official Journal L 126, 08/05/2001, 18-19.

DÍAZ, O., FERNÁNDEZ, M., GARCÍA DE FERNANDO, G.D., HOZ, L. & ORDÓÑEZ, J.A. (1992). Effect of the addition of the aspartyl proteinase from *Aspergillus oryzae* on dry fermented sausage proteolysis during ripening. Proc. 38th Int. Con. Meat Sci. Technol., Clermont-Ferrand. Vol. 4, pp. 779-782.

FLATH, P.A., SUGISAWA, H. & TERANISHI, R. (1981). Introduction-Problems in flavour Research. In *Flavor Research: Recent advances.* R. Teranishi, P.A. Flath and H. Sugisawa (eds.). Marcel Dekker, New York, pp. 1-14.

GARCÍA-REGUEIRO, J., A., GIBERT, J. & DÍAZ, I. (1994). Determination of neutral lipids from subcutaneous fat of cured ham by capillary gas chromatography and liquid chromatography. J. Chromatogr. A, 667, 225-233.

HANSON S.W.F. & OLLEY, J. (1963). Application of the method of lipid extraction to tissue homogenate. Biochem. J. 89, 101P-102P.

JOHNSON, M.J. 1940. Isolation and properties of a pure yeast polypeptidase. Proc. 3rd Int. Cong. Microbiol. New York, p. 348.

MARRA, A.I., SALGADO, A., PRIETO, B. & CARBALLO, J. (1999). Biochemical characteristics of dry-cured lacón. Food Chem. 67, 33-37.

MARTÍN, L., ANTEQUERA, L., RUÍZ, J., CAVA, R., TEJEDA, J.F. & CÓRDOBA, J.J. (1998). Influence of processing conditions of Iberian ham on proteolysis during ripening. Food Sci. Technol. Int. 4, 17-22.

MORRISEY, P.A., SHEELY, P.J.A., GALVIN, K., KERRY, J.P. & BUCKLEY, D.J. (1998) Lipid stability in meat and meat products. Meat Sci. 49, suppl. 1, S73-S86.

SANDERSON, A., PEARSON, A.M. & SCHWEIGERT, B.S. (1966). Effect of cooking procedure on flavor components of beef: carbonyl compounds. J. Agric. Food Chem. 14, 245-250.

SPANIER, A.M., EDWARDS, J.V. & DUPUY, H.P. (1988). The warmed-over flavor process in beef: a study of meat proteins and peptides. Food Tech. 42, 110-118.

TOLDRÁ, F. (1998). Proteolysis and lipolysis in flavour development of dry-cured meat products. Meat Sci. 49, suppl. 1, S101-S110.

WHITFIELD, F.B. (1992). Volatiles from interactions of Maillard reactions and lipids. Crit. Rev. Food Sci. Nutr. 31, 1-58.

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Table 1. Lipid fractions composition (mean ± S.D.) of internal an external muscles from dry cured foreleg (n=5) and cooked dry cured foreleg (n=5)

	Internal muscles			External muscles		
	Dry-cured foreleg	Cooked foreleg	p	Dry-cured foreleg	Cooked foreleg	p
Glycerides (%)	75.1 ± 1.16	70.36 ± 4.44	n.s.	74.23 ± 1.97	71.85 ± 3.72	n.s.
Free Fatty Acids (%)	12.27 ± 1.73	18.29 ± 3.30	*	11.4 ± 1.46	16.35 ± 2.33	**
Phospholipids (%)	12.63 ± 1.13	11.35 ± 2.94	n.s.	14.17 ± 0.63	11.79 ± 2.60	n.s.

n.s.: no significant differences ($p>0.05$)

* $p<0.05$; ** $p<0.01$; *** $p<0.001$

Table 2. Nitrogen fractions composition (mean ± S.D.) in dry cured forelegs (n=5) and cooked dry cured forelegs (n=5) (mg N/g total nitrogen)

	Internal muscles			External muscles		
	Dry cured foreleg	Cooked foreleg	p	Dry cured foreleg	Cooked foreleg	p
NPN	99.34 ± 18.04	74.11 ± 19.85	n.s.	75.36 ± 22.81	53.30 ± 7.03	n.s.
PTN	44.32 ± 16.95	20.51 ± 12.75	*	29.90 ± 16.19	16.25 ± 8.65	n.s.
SSN	33.10 ± 8.11	18.08 ± 1.17	**	21.87± 7.56	14.72 ± 3.58	n.s.
Ammonia	1.83 ± 0.98	6.32 ± 1.11	***	2.36 ± 1.48	5.01 ± 2.95	n.s.

n.s.: no significant differences ($p>0.05$)

* $p<0.05$; ** $p<0.01$; *** $p<0.001$