



# Meat quality of tenderloin from Iberian pigs as affected by breed strain and crossbreeding

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## ABSTRACT

In 2007, a new National Quality Standard was published in Spain to regulate the products derived from the Iberian pig carcass, including for the first time fresh meat. In the same way, four different Iberian strains were recognized as official (Lampião, Entrepelado, Retinto and Torbiscal). A batch ( $n = 10$ ) of each pig strain was selected using neutral DNA markers, and another batch of the most common crossbreeding pigs (Iberian  $\times$  Duroc) was included into the study as a control. The main meat quality parameters of tenderloin, the most expensive meat cut for fresh consumption, from those five pig groups were analysed. Retinto and Lampião strains showed the closest phenotypic distances, followed by Entrepelado strain. Meat from crossed and Torbiscal pigs had lower water holding capacity,  $L^*$  and  $a^*$ , and higher SFA than meat from the other three strains. Crossbred pigs had the lowest protein, intramuscular fat and PUFA contents.

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## 1. Introduction

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 highly priced cured products. However, the importance of the consumption of fresh meat has recently increased, since IB pork is considered as a high quality alternative to meat from lean pigs commonly consumed in Spain (Ramírez & Cava, 2007). Due to this increasing consumption of fresh meat, in November 2007, a National Quality Standard (NQS) was published to regulate production, processing and marketing of products derived from the IB pig carcass (RD 1469/2007). That NQS included for first time fresh meat, instead of cured products only.

In 2007, four IB strains (LA, Lampião; EN, Entrepelado; RE, Retinto and TO, Torbiscal) were recognized as official by the Spanish Ministry of Agriculture, under the Official List of Livestock Breeds (APA/53/2007). The traditional differentiation of the IB strains has been based on morphologic parameters and pedigree-based inbreeding (Fernández, Rodríguez, Toro, Rodríguez, & Silió, 2002). However, no genetic characterization has been done to classify the IB pig populations within each official strain. Moreover the previous NQS (RD 1083/2001) regulated as well in the percentage of IB  $\times$  Duroc crossbreeding allowed for products labelled as “Iberian”, the bulk of the production marketed today.

Only a few studies compared objective fresh meat quality of IB pig strains and “white pig” crossbreeding under semi-extensive

production conditions (Muriel, Ruiz, Ventanas, et al., 2004). Those studies analyzed muscles as *longissimus dorsi*, usually assigned for curing, or *masseter*, lowly priced as compared to tenderloin (m. *psoas major*). Some recent studies focused on IB pig meat (Morcuende, Estevez, Ramirez, & Cava, 2007) have used the tenderloin due to the importance of this muscle in the Spanish fresh meat market.

The aim of this study was to compare the quality traits of the tenderloin, the most expensive meat cut for fresh consumption, of the four purebred strains of IB pig breed. Those results were compared to those from 50% Duroc pigs, the most common crossbreeding used in Spain for IB pig production.

## 2. Materials and methods

### 2.1. Genetic selection

In order to verify the strain genetic purity of the selected animals (LA, Lampião; EN, Entrepelado; RE, Retinto and TO, Torbiscal), a preliminary study was conducted in four herds ascribed to the Studbook. This study was carried out on 140 animals (35 assigned to each official strain according to their morphology) from the Spanish Association of IB Pig Breeders (AECERIBER). A batch of 35 IB (no assigned to any official strain)  $\times$  Spanish Duroc crossbred pigs was included into the study to compare their genetic and productive traits to those from IB purebred pigs. The Spanish Duroc line was selected because it is currently used by the Spanish industry (Serrano, Valencia, Nieto, Lázaro, & Mateos, 2008).

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Blood samples from the animals were collected using Vacutainer® blood collection tubes with EDTA  $k_3$  as anticoagulant. Genomic DNA was isolated and purified using Dominion mbl Kit® (Dominion, Cordoba, Spain). Microsatellite markers (34), including the 27 recommended by FAO for genotyping of pig populations (FAO, 2004), were amplified by polymerase chain reaction (PCR), using a programmable thermal cycler (Mastercycler EP Gradient®, Eppendorf, Germany). The microsatellite markers used were: S0068, S0090, S0101, S0155, S0178, S0215, CGA, IGF1, S0002, S0005, S0026, S0225, S0226, S0227, S0228, S0355, S0386, SW122, SW24, SW240, SW632, SW72, SW857, SW911, SW936, SW951, S0071, S0106, SW1057, SW1111, SW210, SW2419, SW787 and SW874 (US pig gene mapping program, 2007).

PCR products were subjected to electrophoresis in an automatic sequencer ABI 3130 (Applied Biosystems, Foster City, CA, USA), and their results were analyzed with *GeneScan v.3.7* software. Alleles were determined with *GeneMapper 4.0* software. *Genetix 4.05* software (Belkhir, Borsa, Chikhi, Raufaste, & Bonhomme, 2001) was used to compute the allelic frequencies in order to estimate different parameters of genetic variability as expected ( $H_e$ ) and observed ( $H_o$ ) heterozygosities and *Wright's F-Statistics* ( $F_{IS}$ ,  $F_{ST}$  and  $F_{IT}$ ). The  $F_{IS}$  parameter is a fixation index of individuals with respect to subpopulations and it is used to estimate the inbreeding caused by the subdivision of the population and the type of crossbreeding (the coefficients due to the existence of subdivision population and/or mating design). The  $F_{ST}$  parameter is a measure of genetic diversity among populations and shows the rate of genetic differentiation among them while the  $F_{IT}$  parameter is used to measure the absence or excess of heterozygote in the population as a whole. Similarly, a Factorial Correspondence Analysis (FCA) in a three-dimensional scale (Benzecri, 1973) was carried out on individual samples and graphically represented the patterns of distribution of genetic variability based on microsatellite data (individual multilocus scores). Finally, a study of genetic distances among those subpopulations was carried out using Reynolds distance (Reynolds, Weir, & Cockerham, 1983). This is considered as the best measure of genetic distance for studying populations that have recently diverged, as IB strains. To obtain a graphical representation of the distance matrix (Reynolds distance), a tree of genetic distances with the UPGMA algorithm was generated using PHYLIP 3.65 software package (Felsenstein, 2005).

## 2.2. Animal management

From the groups selected in the genetic study, 50 castrated male pigs were used for the meat quality study, 10 from each IB pig strain and 10 from IB  $\times$  Duroc crossbreeding.

All pigs were reared under regular semi-extensive management. IB and crossbreed piglets were born in different farms, but transferred after post-weaning to the same barn. Piglets were weaned at 49–56 days of age, and fattening started at an age of about 12–13 weeks (live weight of 20–23 kg for IB and 24–26 kg for crossbreed pigs). Between weaning and transfer to the farm, IB and crossbreed piglets were fed a commercial starter concentrate. On-farm management practices were identical for all pigs. The pigs were fed *ad libitum* using a cereal-based diet (crude protein: 10.8%; crude fibre: 6.4%; crude fat: 3.5%; total lysine: 0.8%).

## 2.3. Sampling and carcass analysis

The pigs were slaughtered when reached the commercial live weight (160–180 kg; 12 months of age). The animals were transported to the abattoir the day before slaughtering, without mixing pigs from different groups at any moment, and trying to minimize the stress of the animals. Carbon dioxide was used for stunning just before bleeding, according to the specifications outlined in the

Spanish legislation. The pH was measured 24 h after slaughter in tenderloins, using a penetrating electrode adapted to a hand-held Crison pH/mv-506 m.

Tenderloins were collected 24 h *post-mortem* from the left side of the carcass and, after ageing for 72 h at 4 °C, they were analyzed or frozen at 80 °C for future texture and fatty acid analysis.

## 2.4. Meat quality analysis

Percentages of moisture, ash, fat, and protein in the samples were determined according to Association of Official Analytical Chemists – AOAC (1990) methods. Protein content was measured by the block digestion method (UNE 55-020), moisture content was determined by drying at 102 °C for 24 h (ISO R-1442) and ashing was done at 550 °C for 24 h (ISO R-936). The intramuscular fat (IMF) percentage was measured according to Soxhlet method (ISO R-1443) using a Foss Tecator AB Soxtec 2050.

Muscle brightness and colour indices were recorded on fresh meat after 60 min blooming with a chromameter Minolta CM-2500d (illuminant D65; viewing angle 10°). The  $L^*$  (lightness),  $a^*$  (redness) and  $b^*$  (yellowness) values were recorded from the average of three random readings across each muscle surfaces. The concentration of myoglobin was assayed from the total content of heme pigment according to Hornsey (1956), and calculated by multiplying heme pigment concentration by the factor 0.026.

Water holding capacity (WHC) was determined as percentage of free water (Grau & Hamm, 1953). Before texture analysis, meat was thawed under tap water to an internal temperature of 16–18 °C. Meat was cooked in a water bath at 75 °C until the internal temperature reached 70 °C. Shear force (WBSF) was measured on samples 1 cm wide  $\times$  1 cm high. The cores were sheared perpendicular to the muscle fibres orientation using a TA XT2 texture analyser (Stable Microsystems, UK) equipped with a Warner-Bratzler device and crosshead speed set at 2 mm/s.

Total fatty acids were extracted, methylated and analyzed by an adaptation of the method described by Aldai, Osoro, Barron, and Nájera (2006), which has been reported to be highly effective for PUFA analysis (Juárez et al., 2008). Separation and quantification of the fatty acid methyl esters was carried out using a gas chromatograph (GC, Agilent 6890N, Agilent Technologies Spain, S.L., Madrid, Spain) equipped with a flame ionisation detector automatic sample injector HP 7683, and using a HP-88 J&W fused silica capillary column (100 m, 0.25 mm i.d., 0.2  $\mu$ m film thickness, Agilent Technologies Spain, S.L., Madrid, Spain). Nonadecanoic acid methyl ester (C19:0 ME) at 10 mg/ml was used as an internal standard. Individual fatty acid methyl esters were identified by comparing their retention times with those of an authenticated standard fatty acid mix Supelco 37 (Sigma Chemical Co. Ltd., Poole, UK). Identification of *cis9-trans11* CLA isomer, was achieved by comparing its retention time with that of an authenticated standard from Matreya (>98% purity; Matreya, LLC, Pleasant Gap, USA). Fatty acids were expressed as a percentage of total fatty acids identified and grouped as follows: SFA, MUFA and PUFA. PUFA/SFA and n-6/n-3 ratios were calculated.

## 2.5. Statistical analysis

Meat quality data were analyzed with the Statistica 7.0 for Windows (StatSoft, 2006) statistical package. A general lineal model was used to determine the significance of the effects of the different strains and crossbreeding on meat quality traits. Carcass weight was fitted as a lineal covariate. A tree diagram (cluster analysis) was developed following Ward's method to show the distribution of the five groups of animals according to their meat quality traits.

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