

Racial verification of Iberian ham and shoulders commercialized in Spanish supermarkets

Caraballo, C.^{1,2}; Muñoz, M.^{1,2@}; Rodríguez, C.²; Silió, L.² and García-Casco, J.M.^{1,2}

¹Centro de I+D en Cerdo Ibérico, INIA, Zafra, Spain.

²Departamento de Mejora Genética Animal, INIA, Madrid, Spain.

ADDITIONAL KEYWORDS

Dry-cured products.
Traceability analyses.
SNP chip.
Quality standard.

SUMMARY

The commercialization of Iberian meat and dry-cured products is regulated by the Spanish law. A first decree approved on 2007 (Real Decreto 1469/2007) established that Iberian products designated as "Iberian" have to come from pigs with at least a 50% of their genome from Iberian breed and the remaining percentage from Duroc. This decree was modified on 2014 (Real Decreto 4/2014) and it requires that, to be labelled as "100% Iberian", products must come from purebred pigs and maintains the previous regulation about "Iberian" label. A traceability SNP chip containing 64 SNPs was developed by the Pig Breeding and Genetics Group of INIA. It allows discriminating between the proportion of Iberian and Duroc genomes of an individual. The objective of this study was to check if the dry-cured products (hams and shoulders) sold as "Iberian" by some of the major distributors of Iberian products at main Spanish supermarkets follow the law. One hundred and sixteen samples of dry-cured ham and shoulder sliced packs commercialized as "Iberian" were genotyped with the chip. The genotyping data was analysed using BAPS 5.3. A percentage of Iberian genome lower than 40% was estimated for 34.5% of the samples, which do not follow the regulation of genetic origin. It is worth to note that five of the 116 samples were 100% Duroc. None of the suppliers included in the study met the law strictly. These results show that, in general, the Quality Standard has not been obeyed for a long time. A future new sampling will show if the stricter controls of the animals set by the 2014 law has been useful to improve the legal compliance.

Verificación racial de jamones y paletas Ibéricos comercializados en supermercados españoles

RESUMEN

La Norma de Calidad española regula la comercialización tanto de producto fresco como curado procedente de raza Ibérica. La primera Norma, que fue aprobada en 2007 (Real Decreto 1469/2007), establecía que los productos etiquetados como "Ibérico" debían proceder de cerdos con al menos un 50% del genoma Ibérico y que el porcentaje restante debía proceder de Duroc. Esta Norma fue modificada en 2014 (Real Decreto 4/2014) añadiendo que el producto etiquetado como "100% Ibérico" debe proceder de cerdos Ibéricos puros y mantiene la misma regulación que en 2007 para el producto etiquetado como "Ibérico". En el Grupo de Genética de Cerdos del INIA se ha desarrollado un chip de 64 SNPs para trazabilidad genética, que permite inferir la proporción del genoma de origen Ibérico y Duroc de animales y productos cárnicos. El objetivo de este trabajo consistió en comprobar si los productos curados (jamones y paletas) etiquetados como "Ibérico" por fabricantes y distribuidores representativos del sector y comercializados en los principales supermercados españoles cumplen este aspecto de la Norma de Calidad. 116 muestras problema procedentes de paquetes de loncheados de jamón y paleta comercializados como "Ibéricos" fueron genotipadas con este chip. Los genotipos fueron analizados usando el programa BAPS 5.3. En el 34.48% de las muestras analizadas la proporción estimada de genoma de origen Ibérico era inferior al 40% incumpliendo claramente la Norma. Cabe destacar que en cinco de las 116 muestras la proporción estimada de genes Duroc era del 100%. Estos resultados son indicativos de un insatisfactorio cumplimiento de la Norma de Calidad en diez de los doce distribuidores analizados. Un muestreo futuro mostrará si los controles más estrictos impuestos con la nueva Norma de 2014 han servido para mejorar el cumplimiento de la ley.

PALABRAS CLAVE ADICIONALES

Productos curados.
Análisis de trazabilidad.
SNP chip.
Norma de Calidad.

INFORMATION

Cronología del artículo.
Recibido/Received: 13.01.2017
Aceptado/Accepted: 06.07.2017
On-line: 15.01.2018
Correspondencia a los autores/Contact e-mail:
aparicio@unex.es

INTRODUCTION

Iberian pigs have been traditionally crossed with individuals of Duroc breed for improving lean growth efficiency and the percentage of carcass premium cuts.

Spanish law regulates these crosses and the commercialization and labelling of their meat and dry-cured products. The Spanish Royal Decree (RD 1469/2007) established that Iberian products designated as "Iberian" have to come from the progeny of Iberian dams

crossed with Duroc sires. This decree was modified on 2014 (RD 4/2014) imposing stricter controls in the productive chain to avoid potential frauds and it also requires the products must to be labelled as “100% Iberian” when they were obtained from purebred pigs. During the last years, several approaches have been carried out to discriminate between Iberian and Duroc alleles using different genetic markers. A panel of 36 microsatellites allowed discriminating between Iberian and Duroc breeds (Fabuel et al. 2004, p. 104-13), however, the efficiency of these markers to differentiate between purebred and 50% or 75% crossbred individuals has been questioned (Rodríguez-Ramilo et al. 2010, p. 347-355). In addition to this, microsatellites are scarcely useful as diagnostic markers since breed-specific alleles are not common and they have a low allelic frequency (Barragan et al. 2007). Although SNPs are usually bi-allelic and less informative than microsatellites, they are more advantageous since they appear in a higher abundance in the genome, it is easier to carry out an interpretation and automation is more readily implemented. Different SNP panels have been proposed up to now for this goal: private alleles of the Iberian or Duroc breeds identified in some genes (Fernández et al. 2004, p. 1855-1860), mitochondrial markers (Alves et al. 2009, p. 1216-23) and a 96 SNP panel with divergent allelic frequencies between Iberian and Duroc pigs (Muñoz et al. 2013). This last panel verified successfully the breed origin of Duroc x Iberian crossbred pigs, but it had several inconveniences, including the cost and lower than expected differences in allelic frequencies between Duroc and Iberian. Barragan et al. (2015) improved the previous panel of 96 SNPs, designing a new panel with 64 SNPs which is able to differentiate between the proportion of Iberian and Duroc alleles of an individual and with a lower cost. The objective of the current work consisted in using this panel to check if the dry-cured products (hams and shoulders) sold as “Iberian” by some of the major commercial distributors at main Spanish supermarkets follow the law.

MATERIAL AND METHODS

SAMPLING

A total of 116 ham and shoulder sliced packs commercialized as “Iberian” were selected from 12 different suppliers. Ten packs were sampled per supplier, except for distributor 1 with six packs sampled. These packs were sold in eight different supermarkets located in five different regions of Spain: Madrid, Extremadura, Castilla La-Mancha and Andalucía. The packs were purchased along two years to avoid sampling packs belonging to the same production batch.

GENOTYPING AND ADMIXTURE ANALYSIS

Genomic DNA samples were extracted using the NucleoSpin® Food kit (Macherey-Nagel). DNA samples from the 116 samples and 245 reference samples with a known genetic origin (138 Iberian 100%, 86 Duroc 100%, 12 Iberian 50% and 9 Iberian 75%) were genotyped for an SNP panel with 64 SNPs (Barragan et al. 2015) using Taqman OpenArray™ genotyping plates custom-designed in a QuantStudio™ 12K flex Real-Time PCR System (ThermoFisher Scientific).

An admixture analysis was carried out with the genotyping data of the 62 SNPs using BAPS 5.3 software since two out of the 64 SNPs failed during the genotyping process.

RESULTS

The samples were classified in four categories according to their estimated percentage of Iberian genome: ≥ 60 , 50-60, 40-50 and < 40 . **Table I** shows the percentage of samples allocated to each category. A 13.8% and a 40.5% of the samples show percentages of Iberian genome of 50-60 and 50-40, respectively, and could be accepted as being in accordance with the Spanish Quality legislation. However, 34.5% of the samples present a percentage of Iberian genome lower than 40% and clearly exhibit an inadequate labelling.

The percentage of samples per category of each distributor was uneven. The **Figure 1** shows a relevant heterogeneity intra- and inter-distributor. All the samples of distributor 7 have a percentage of Iberian genome above 40%. On the other hand, this percentage is lower than 40% in all the samples of distributor 8, and any of the samples would not follow the Spanish Standard Quality. Moreover, five distributors supply ham packs of astonishing heterogeneous racial origin, allocated to the four categories distinguished in this study (Distributors 4, 5, 6, 9 and 10).

DISCUSSION

In standard crossbreeding schemes, only levels of 50 or 75 percent of Iberian genomes would be expected in “Iberian” labelled products. The current study shows deviations of these figures in the percentages of Iberian genome found in the samples analyzed. It may be attributed to the likely introgression of Duroc into the genome of some Iberian reproducers, not detected by the morphological identification made by specialized technicians in the farm. Therefore, an important proportion of the samples analysed have been inappropriately labelled as “Iberian”. The consumer confidence on the labelling may be also questioned by the observed heterogeneity intra- and inter-distributors. However, some of them do a tolerable compliance of the regulation (**Figure 1**). The new law RD 4/2014 improves the controls carried out along the productive chain. Therefore, we expect a better fulfilment of the

Table I. Percentage of marketed samples of “Iberian” sliced hams and shoulders allocated to four categories of different proportion of Iberian genome (Porcentaje de muestras comercializadas en lonchas de jamones y paletas “ibéricos” asignadas a cuatro categorías de diferente proporción del genoma ibérico).

% Iberian genome	% Samples ($n = 116$)
≥ 60	11.21
50 – 60	13.79
40 - 50	40.52
< 40	34.48

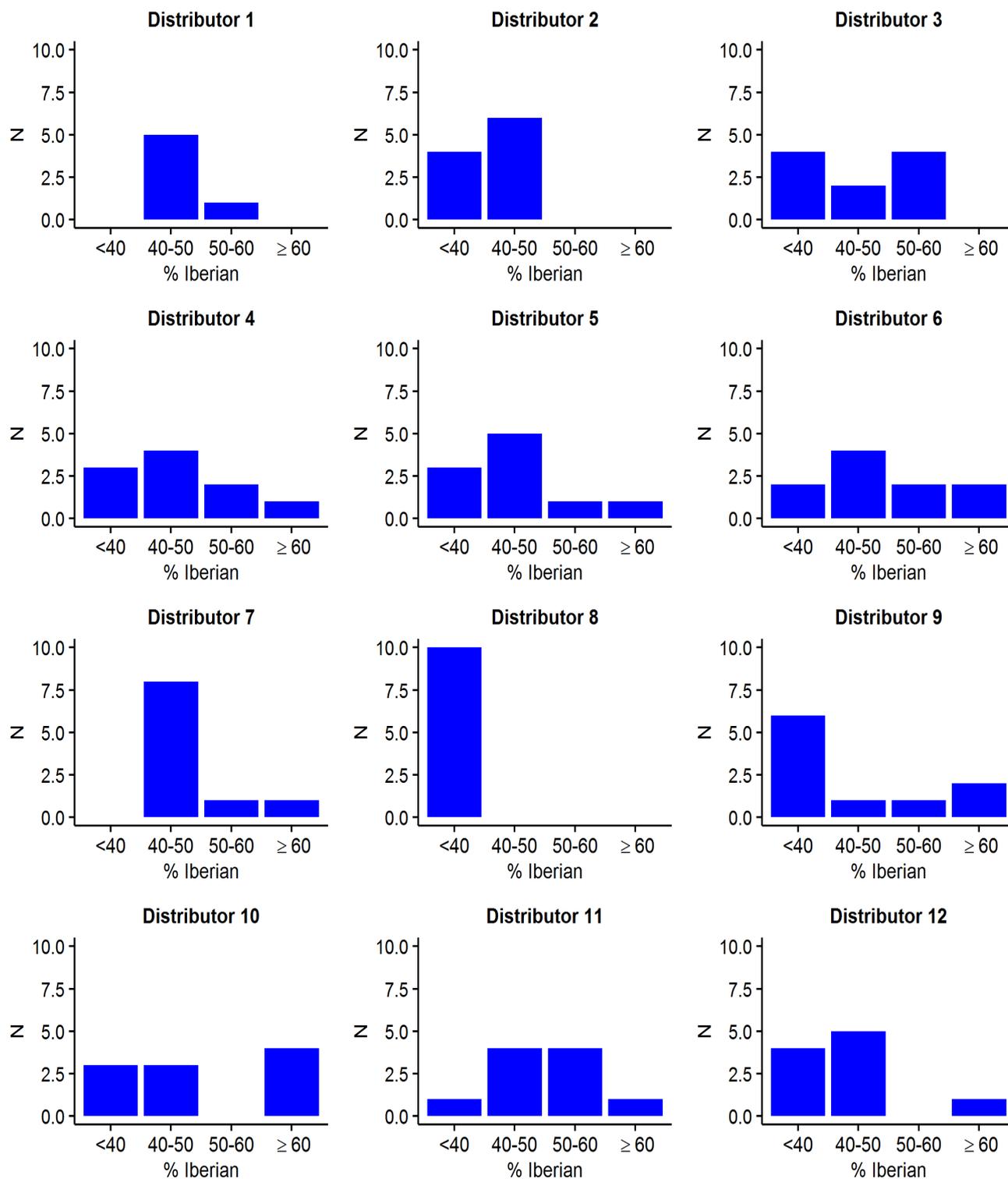


Figure 1. Number of samples per category within each of the 12 distributors analyzed (Número de muestras por categoría en cada uno de los 12 distribuidores analizados).

law with a new sampling performed with dry-cured products from animals qualified under the new law.

In simulations made by Barragán et al. (2015), type 1 error values of 0.019 and type 2 error values of 0.018 were achieved with this panel for assignment of Duroc x Iberian F1 animals. However, in the practical, the ability to solve problems is lower than the expected

in the simulations since two markers did not work and there were not data for some markers due to genotyping failures. An improvement of the SNP panel, including new SNPs with more extreme differences of allelic frequencies will allow a more precise estimation of the percentage of Iberian and Duroc genomes in the problem samples.

BIBLIOGRAPHY

- Alves, E, Fernandez, A I, Fernandez-Rodriguez, A, Perez-Montarelo, D, Benitez, R., Ovilo, C, Rodriguez, C & Silio, L 2009, 'Identification of mitochondrial markers for genetic traceability of European wild boars and Iberian and Duroc pigs', *Animal*, vol. 3, pp. 1216-23.
- Barragan, C, Garcia Casco, J, Garcia-Cortes, L, Garcia, F, Nuñez, Y, Rodriguez, C & L., S 2015, 'Refining SNPs-based tests for verifying the raw material of meat products from crossbred and purebred Iberian pigs', *VIIIth Dry Cured Ham World Congress*. Toulouse.
- Barragan, C, Rodriganez, J, Garcia Casco, J, Martín-Palomino, P & Mc, R 2007, 'Validation of microsatellite markers for routine parentage testing in Iberian pigs', *6th International Symposium on the Mediterranean Pig*. Capo d'Orlando.
- Fabuel, E, Barragan, C, Silio, L, Rodriguez, M C & Toro, M A 2004, 'Analysis of genetic diversity and conservation priorities in Iberian pigs based on microsatellite markers', *Heredity*, vol. 93, pp. 104-13.
- Fernández, A, Fabuel, E, Alves, E, Rodriguez, C, Silió, L & Óvilo, C 2004, 'DNA tests based on coat colour genes for authentication of the raw material of meat products from Iberian pigs', *Journal of the Science of Food and Agriculture*, vol. 84, pp. 1855-60.
- Muñoz, M, Garcia Casco, J, Caraballo, C, Rodriguez, M, Barragan, C, Alves, E & Silio, L 2013, 'Autenticacion del origen racial iberico puro o cruzado mediante un panel de marcadores SNPs', *VII Congreso Mundial del Jamón*. Ourique.
- Rodriguez-Ramilo, S T, Toro, M A & Fernandez, J 2010, 'Usefulness of the clustering methodologies to discriminate between purebred and crossbred individuals', *Spanish Journal of Agricultural Research*, vol. 8, pp. 347-55.