

POSTER

NEGRO DE LOS PEDROCHES, THE MOLECULAR DEFINITION OF A NEW VARIETY OF THE IBERIAN PIG BREED

NEGRO DE LOS PEDROCHES, LA DEFINICIÓN MOLECULAR DE UNA NUEVA VARIEDAD DE LA RAZA PORCINA IBÉRICA

Martínez, A.M.¹, J.V. Delgado¹, J.L. Vega-Pla², F. Escribano³ and A. Cabello⁴

¹Departamento de Genética. Campus de Rabanales. Ctra. Madrid-Cádiz, km. 396. 14071 Córdoba. España.

²Laboratorio de Grupos Sanguíneos. Cría Caballar. Córdoba. España.

³Asociación Guadamatilla. Pozoblanco. Córdoba. España.

⁴Delegación de Investigación y Desarrollo Agrícola y Ganadero. Diputación de Córdoba. Palacio de la Merced, s/n. 14071 Córdoba. España.

ADDITIONAL KEYWORDS

Microsatellites. Conservation.

PALABRAS CLAVE ADICIONALES

Microsatélites. Conservación.

SUMMARY

Pedroches Valley is one of the most important areas of the traditional pig breeding in Andalusia (Southern Spain). The Iberian pig variety named Negro de Los Pedroches has its origins in this valley. This is an old autochthonous pig population generated after decades of breeding but now in risk of extinction. Breeders consider this Iberian pig strain as an important element of the Andalusian paddock linked to its cultural and biological patrimony. It seems to have excellent productive capabilities that justify its characterisation and conservation. Microsatellites are used to investigate the genetic relationships with other Iberian pig subpopulations, the use of an accurate panel of loci increases the chance of their definition. Genetic distances suggest that the Negro de Los Pedroches must be considered as another variety.

de Los Pedroches, tiene su origen en el valle de Los Pedroches (Andalucía, sur de España). Es una antigua población autóctona generada después de décadas de cría pero que ahora está en riesgo de extinción. Los criadores consideran a esta estirpe como un elemento importante de la dehesa andaluza ligado a su patrimonio cultural y biológico. Parece tener excelentes capacidades productivas que justifican su caracterización y conservación. Los microsatélites son usados para investigar las relaciones genéticas con otras subpoblaciones del cerdo Ibérico, el uso de un panel efectivo de *loci* incrementa la oportunidad de su definición. Las distancias genéticas muestran como el Negro de Los Pedroches debe ser considerado como otra variedad de la raza porcina Ibérica.

RESUMEN

La variedad de cerdo Ibérico llamada Negro

INTRODUCTION

The European pig genetic resources are essentially formed from breeds

Arch. Zootec. 52: 219-223. 2003.

belonging to two ancient strains: the Celtic type in north-central Europe and the Mediterranean type in the south (Porter and Tebbit, 1993). Both types were represented in Spain by native breeds, but today only the Iberian Pig from Mediterranean type is completely developed. Other autochthonous breeds are rare, endangered or extinct.

The Iberian Pig is among the most important livestock of Spain, bred mainly in traditional extensive (sustainable) conditions and benefiting important ecosystems such as the Mediterranean paddock named *dehesa*, which produces high quality derived products (loin, ham etc) with high profitability. Classically the Iberian Pig breed was divided into several varieties based in

morphological, productive traits and more recently using molecular markers (Martínez *et al.*, 2000).

The Negro de Los Pedroches (**figure 1**) is an ancient variety of the Iberian Pig from the Pedroches Valley (Province of Córdoba, Spain) that has remained until the present times, although today it is in great risk of extinction. The Negro de Los Pedroches pig has certain characteristics that make it unique and worthy for conservation: it has a typical morphology of the Iberian Pig and a good reproductive ability, meat/carcass conformation and weight gain performance. The project to rescue this variety includes a genetic analysis using 25 microsatellites recommended by



Figure 1. Female of the Negro de Los Pedroches Variety. (Hembra de la variedad Negro de Los Pedroches).

MOLECULAR DEFINITION OF A NEW IBERIAN VARIETY

Table 1. *Nei Da genetic distances between pairs of populations.* (Distancias genéticas Nei Da entre pares de poblaciones).

| | Duroc | Dorado | Entrep | Lampiño | Manchado | Pedroc | RExtrem | RPortu | Silvela |
|-------------------|-------|--------|--------|---------|----------|--------|---------|--------|---------|
| Dorado Gaditano | 0.256 | | | | | | | | |
| Negro Entrepelado | 0.253 | 0.108 | | | | | | | |
| Negro Lampiño | 0.292 | 0.204 | 0.171 | | | | | | |
| Manchado Jabugo | 0.349 | 0.305 | 0.175 | 0.293 | | | | | |
| Negro Pedroches | 0.291 | 0.213 | 0.179 | 0.197 | 0.282 | | | | |
| Retinto Extremeño | 0.283 | 0.130 | 0.031 | 0.161 | 0.178 | 0.199 | | | |
| Retinto Portugués | 0.311 | 0.219 | 0.125 | 0.237 | 0.259 | 0.283 | 0.139 | | |
| Silvela | 0.297 | 0.131 | 0.066 | 0.162 | 0.218 | 0.176 | 0.069 | 0.132 | |
| Torbiscal | 0.411 | 0.272 | 0.195 | 0.187 | 0.280 | 0.289 | 0.190 | 0.244 | 0.144 |

ISAG-FAO for studies on porcine biodiversity. In spite of not showing high values of genetic diversity, it can be stated that the Negro de los Pedroches pig is a breed variety that perfectly fits the profile of the Iberian Pig breed, together with other well-defined varieties.

The objective of this work was to estimate if the Negro de Los Pedroches variety belongs to the Iberian Pig breed, based on genetic distance analysis based on microsatellite markers and if its particular genotypic profile can be considered sufficiently different to be considered another variety of the Iberian Pig.

MATERIAL AND METHODS

A sample of 22 individuals of Negro de Los Pedroches pig was analysed, selected according to their genetic characteristics and grouped at a ranch of the Guadamatilla Association in Pozoblanco (Córdoba). DNA was extracted from blood samples using

the Kawasaki method (Kawasaki, 1990). The microsatellites used for the analysis were: *CGA*, *IGF1*, *S0002*, *S0005*, *S0026*, *S0068*, *S0090*, *S0101*, *S0155*, *S0178*, *S0215*, *S0225*, *S0226*, *S0227*, *S0228*, *S0355*, *S0386*, *SW122*, *SW24*, *SW240*, *SW632*, *SW72*, *SW857*, *SW911*, *SW936* and *SW951*. The microsatellites were amplified using the polymerase chain reaction (PCR) by several multiplex reactions (Martínez *et al.*, 2000). Electrophoresis was carried out using the ABI 377 XL automatic sequencer (Applied Biosystems, Foster City, CA, USA).

The genotypes obtained were compared with those available for other varieties of the Iberian Pig breed (Martínez, 2001). Nei genetic distances were calculated between populations (Nei, 1972) as well as the D_{SA} individual distance based on the proportion of alleles shared for two individuals averaged over loci (Bowcock *et al.*, 1994) using the computer program MICROSAT v.1.5b (Minch, 1997). An UPGMA tree was constructed with

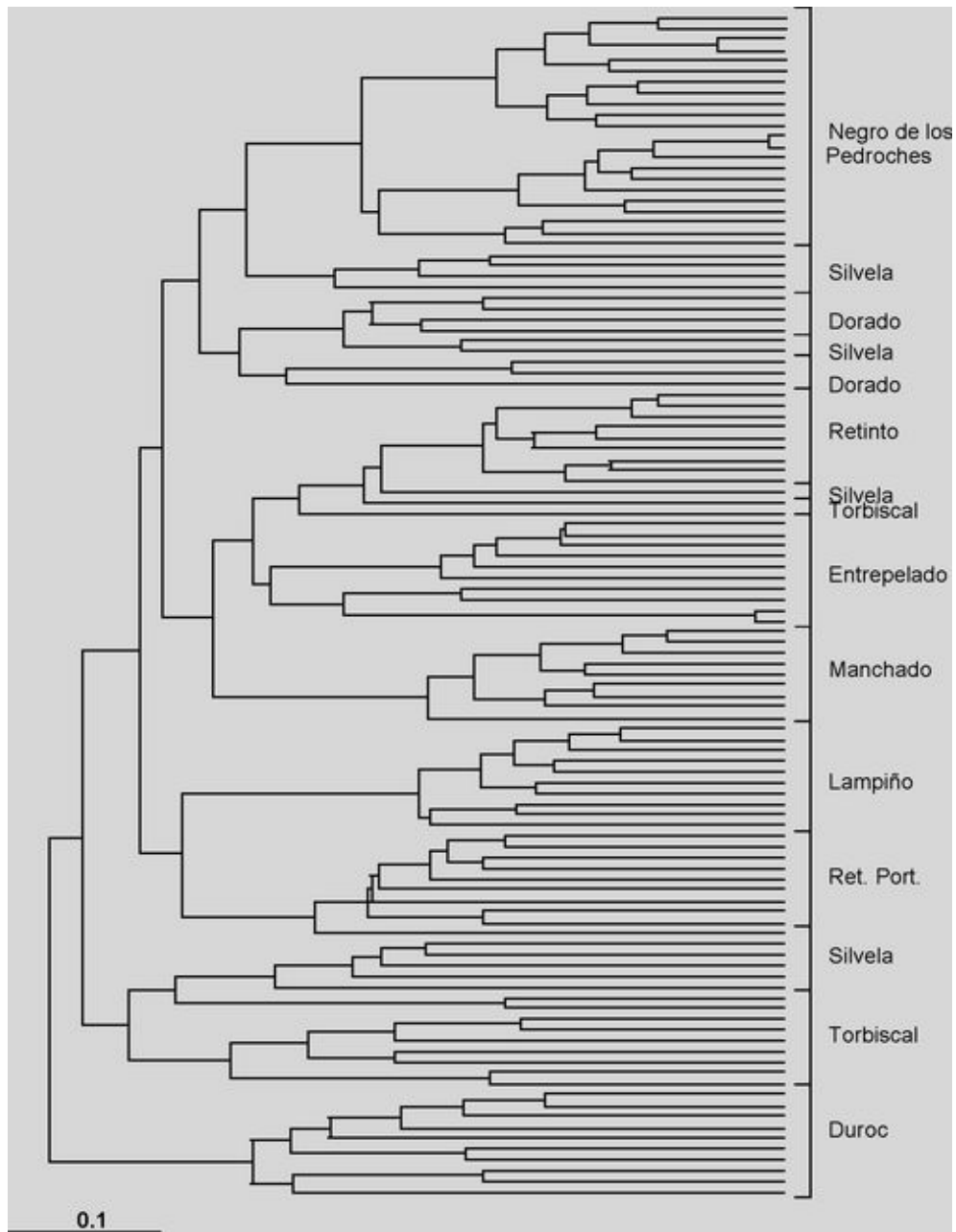


Figure 2. UPGMA tree based on individual genetics distances. (Árbol UPGMA basado en distancias genéticas individuales).

MOLECULAR DEFINITION OF A NEW IBERIAN VARIETY

the D_{SA} values using the NEIGHBOR module of the program PHYLIP v.3.57c (Felsenstein, 1995).

RESULTS

All loci were polymorphic and the number of alleles varied between two and eleven. The average of the observed number of alleles was 4.16, heterozygosity by direct count (H) was 0.538 and expected heterozygosity reach 0.576, these values are similar to those obtained in other Iberian Pig varieties (Martínez *et al.*, 2000)

Twenty Duroc individuals were included as an outgroup for the genetic distance computations and showed the largest distances from all other populations. The smallest genetic distance for Negro de Los Pedroches (**table I**) was with Entrepelado followed with Lampiño, indicating an influence of these varieties in its origins.

The UPGMA individual tree of population samples (**figure 2**) shows a

variety clustering of Iberian Pig with the exception of Silvela which appears mixed in some clusters. Lampiño, Torbiscal, Retinto Portugués, Manchado de Jabugo and Negro de los Pedroches samples were clustered into different groups.

The Negro de Los Pedroches pig shares important genetic, as well as phenotypic, characteristics with the Iberian Pig, as it fits perfectly within the group of varieties of this breed that were analysed here. These results are in concordance with those expected from morphological and productive traits.

ACKNOWLEDGEMENTS

This work was financed by Project RZ00-15 of Programa Nacional de Recursos y Tecnologías Agroalimentarias of Ministerio de Ciencia y Tecnología and developed in the molecular genetics laboratory of Delegación de Investigación y Desarrollo Agrícola y Ganadero of Diputación de Córdoba.

REFERENCES

- Bowcock, A.M., A. Ruiz-Linares, J. Tomfohrde, E. Minch, J.R. Kidd and L.L. Cavalli-Sforza. 1994. High resolution of human evolution with polymorphic microsatellites. *Nature*, 368: 455-457.
- Felsenstein, J. 1995. PHYLIP (Phylogeny Inference Package) Version 3.5c. University of Washington.
- Kawasaki, E. 1990. Sample preparation from blood, cells and other fluids. PCR Protocols: A Guide to Methods and Applications. 146-152. Academic Press Inc., New York.
- Martínez, A.M., J.V. Delgado, A. Rodero and J.L. Vega-Pla. 2000. Genetic structure of the Iberian Pig breed using microsatellites. *Animal Genetics*, 31: 295-301.
- Martínez, A.M. 2001. Caracterización genética del cerdo Ibérico mediante marcadores moleculares. Tesis Doctoral. Universidad de Córdoba.
- Minch, E. 1998. MICROSAT Version 1.5b (Macintosh). University of Standford. Standford.
- Nei, M. 1972. Genetic distance between populations. *American Naturalist*, 106: 283-282.
- Porter, V. and J. Tebbit. 1993. Spain and Portugal. In: Pigs: A Handbook to the Breeds of the World, 137-40. Helm Information Ltd. East Sussex.