

# Genealogical analysis of the Gochu Asturcelta pig breed: insights for conservation

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**ABSTRACT:** The Breeders Association of Gochu Asturcelta (ACGA) initiated a recovery programme with six founders (three boars and three sows) in 2002. Information provided by a total of 3156 records (515 litters, 109 boars, and 309 sows) with father and mother known included in the Gochu Asturcelta herdbook was analyzed. The aim of the analyses was to assess if the mating policy implemented by the breeders association ACGA has been successful in preserving the genetic background of the founders in the present population. Two reference populations were defined: 232 individuals born in 2013 and 2014 (PopO), and 18 individuals born in 2012 and 2013 and kept for reproduction (PopR). Mean inbreeding was very high ( $0.230 \pm 0.078$  for the whole pedigree) with mean individual increase in inbreeding ( $\Delta F_i$ ) of  $0.067 \pm 0.021$  for PopO and PopR. However, mean  $\Delta F_i$  tended to keep steady values (around 0.07) after 2009. The number of founders identified for PopO and PopR was 4 and 5, respectively. The ratio between the effective number of ancestors ( $f_a$ ) and effective number of founders ( $f_e$ ) was roughly 1 for both PopO and PopR. This suggests that the ACGA's mating policy has avoided an extreme bottleneck in the population. However, drift caused noticeable losses of genetic diversity: ratio between the founder genome equivalents ( $f_g$ ) and  $f_e$  was around 0.6 for the two reference populations. The current results highlight the need of unbalancing further genetic contributions to the population towards descendants of a very poorly represented founder (Fema 1).

**Keywords:** genetic variability; pedigree analysis; founder contribution; conservation of genetic stock

## INTRODUCTION

Gochu Asturcelta is an extremely endangered pig breed native of Asturias (northern Spain). The breed derives from the ancient Asturiana pig breed (Aparicio 1944), related to the Galician Celta or the Portuguese Bísara breeds (Alvarez Sevilla 2005), which was one of the most developed domestic pig populations in Spain till the second half of the 20<sup>th</sup> century. The intensification of pig production and the introduction of cosmopolitan pig breeds led to a dramatic decline of population size and the Asturiana pig populations nearly became extinct at the end of the 20<sup>th</sup> century. In 2002, a group of enthusiastic farmers founded the breeders associa-

tion ACGA and initiated a recovery programme with six founders (three boars: 9000 L'Utiru, 9002 Machu, and 9005 Tixu, and three sows: 9001 La Preñá, 9003 Fema 1, and 9006 Fema 2) showing accordance with the ancient type of the breed. In 2005, a government-run herd owned by the regional administration of Asturias was founded at the facilities of SERIDA-Villaviciosa to contribute to the recovery of the breed. Descendants of the ACGA's founders were bred at the herd of SERIDA to supply the Gochu Asturcelta reproductive individuals to interested farmers. At present, the breed is officially included in the Spanish Catalogue of Livestock Breeds (Regulation APA/53/2007) and the Gochu Asturcelta herdbook includes (May

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2014) 4884 individuals registered and 46 herds. Since the beginning of the breeding programme a total of 68 herds became part of the ACGA till August 2014 giving, on average, 72 records each to the herdbook of the breed.

The most usual scenario for livestock populations undergoing conservation programmes is the formation of a founder population, as large as possible, which quickly undergoes a strong population bottleneck due to selection for desirable type characteristics or unexpected reproduction failure for some of the founders (Royo et al. 2007; Alvarez et al. 2008). The Gochu Asturcelta situation departs from this general scenario: the founder population size was extremely small and mating policy aimed at keeping founder contributions balanced across generations. The reproductive career of the direct descendant of the founders was prolonged as much as possible and later the SERIDA's multiplier herd was opened to new reproductive individuals selected from the population according to their type characteristics and low genetic representation. The dependence on the SERIDA's multiplier herd has been further used by the ACGA to give guidelines to the farmers to avoid matings between very close relatives. A decade after triggering the conservation programme of the Gochu Asturcelta pig breed the information included in the herdbook reaches depth enough to assess if the ACGA's mating policy has been successful in preserving the genetic background of the founders in the present population.

Genealogical analyses have proved to be useful to characterize genetic variability and to implement strategies for the preservation of the genetic background (Alvarez et al. 2010; Pjontek et al. 2012). Reports analyzing pedigree data in pig are scant (Toro et al. 2000; Melka and Schenkel 2010; Welsh et al. 2010; Tang et al. 2013). Despite the local origin and development of the breed, the analysis of the very particular genetic scenario of the Gochu Asturcelta pig breed will give insights of general interest for breeders and conservationists.

## MATERIAL AND METHODS

**Data and reference populations.** Data provided by the breeders association (ACGA) included 4883 records belonging to individuals registered in the herdbook from its foundation to August 2014. Non-founder individuals with no genealogies and

no offspring in data (mainly used for slaughtering) were removed from the dataset. A total of 38 records belonging to non-founder individuals with offspring in data did not have identification on father (26) or mother (12). To avoid any noise in the assessment of the breeding patterns, these individuals and their offspring were removed from the analyzed dataset. Therefore, the final dataset included 3156 records from 515 litters. A total of 109 boars and 309 sows had offspring in data.

Two different reference populations were defined for genetic analyses: (1) PopO, including the offspring (232 individuals) born in 2013 and 2014, and (2) PopR, including 18 individuals born in 2012 and 2013 and kept for reproduction. For descriptive purposes, most parameters will be given for the whole pedigree as well.

**Parameters computed.** The following parameters were computed using the program ENDOG v. 4.8 (Gutierrez and Goyache 2005):

Generation intervals were computed, using birth dates of registered animals together with those of their fathers and mothers, as the average age of parents at the birth of their useful offspring. Four pathways (father–son, father–daughter, mother–son and mother–daughter) were considered.

The number of equivalent complete generations traced ( $t$ ) computed as the sum of  $(1/2)^n$ , where  $n$  is the number of generations separating the individual from each known ancestor (Maignel et al. 1996).

The inbreeding coefficient ( $F$ ), defined as the probability that two alleles at a randomly chosen locus are identical by descent (Malecot 1948), and the average relatedness coefficient (AR), defined as the probability that an allele randomly chosen from the whole pedigree belongs to a given animal (Goyache et al. 2003; Gutierrez et al. 2003), were computed for each individual included in the dataset. Note that the AR coefficient of a founder means its genetic contribution to the population. For each reference population, these coefficients can be summed up for the founders to ascertain their relative contributions to the studied population.

The probability of gene origin was characterized by computing the following parameters: (a) effective number of founders ( $f_e$ ), which is the reciprocal of the probability that two alleles drawn at random in the studied population originate from the same founder (James 1972), computed

from the genetic contribution of founders to the descendant gene pool of the population (Lacy 1989); (b) effective number of ancestors ( $f_a$ ), defined as the minimum number of ancestors, not necessarily founders, explaining the complete genetic diversity of a population (Boichard et al. 1997). Parameter  $f_a$  does not fully account for gene loss by drift from the ancestors to a reference population but complements the information offered by  $f_e$  accounting for the losses of genetic variability produced by the unbalanced use of reproductive individuals producing bottlenecks (Boichard et al. 1997; Gutierrez et al. 2005); and (c) the founder genome equivalents ( $f_g$ ) (Ballou and Lacy 1995), defined as the theoretically expected number of founders that would be required to provide the genetic diversity in the present population if the founders were equally represented and had lost no alleles, was obtained by the inverse of twice the average coancestry of the individuals within each reference population (Caballero and Toro 2000). Finally, the effective number of non-founders ( $nf_e$ ) was computed following Caballero and Toro (2000) as

$$nf_e = \left( \frac{1}{f_g} - \frac{1}{f_e} \right)^{-1}$$

where:

$1/f_g$  = estimator of the gene diversity accounting for unequal contributions of founders (GD)

$1/f_e$  = estimator of the gene diversity or expected heterozygosity in the founder population (GD\*)

Therefore,  $1/nf_e$  estimates the genetic diversity lost due to random drift (RD), where

$$RD = GD^* - GD$$

From the coancestry information, Wright's (1969)  $F_{IS}$  statistics was obtained as

$$F_{IS} = \frac{\tilde{F} - \bar{f}}{1 - \bar{f}}$$

where:

$\tilde{F}$  = mean inbreeding coefficient for the entire metapopulation

$\bar{f}$  = average coancestry for the subpopulation (Caballero and Toro 2000, 2002)

Effective population sizes ( $N_e$ ) were estimated on the basis of the individual increase in inbreeding  $\Delta F_i$  (Gutierrez et al. 2009) and coancestry  $\Delta C_{ij}$  (Cervantes et al. 2011a) considering

$$\Delta F_i = 1 - {}^{t_i-1}\sqrt{(1 - F_i)}$$

$$\Delta C_{ij} = 1 - {}^{(t_i+t_j)/2}\sqrt{(1 - C_{ij})}$$

where:

$F_i$  = inbreeding coefficient of individual  $i$

$C_{ij}$  = coancestry coefficient between individuals  $i$  and  $j$

$t_i, t_j$  = respective equivalent complete generations

Finally, effective sizes were computed using the following formulae:

$$N_e F_i = 1/(2 \Delta F)$$

$$N_e C_{ij} = 1/(2 \Delta C)$$

## RESULTS

The edition of the available dataset ensured the quality of the information analyzed: up to 97.2% of the individuals had all grand-fathers known and 80.2% had all great grand-fathers known.

**Demographic parameters.** Figure 1 illustrates the development of the Gochu Asturcelta pig breed herdbook. The maximum number of active herds in data was 35 in 2009 and 2010. However, the number of herds contributing to the herdbook with useful offspring during the same period was 20. After 2010 the number of herds contributing to the herdbook quickly decreased with only 3 herds giving offspring with father and mother known in 2013. The number of individuals, with father and mother known, registered in the herdbook by year increased from its foundation to 2009 (with a maximum of 724 records). The highest number of sows giving offspring with complete genealogies to the herdbook was 98 in 2008. Consistently with the decrease in the number of active herds, only 240 individuals were registered in the herdbook in 2012 with only 18 individu-

Table 1. Mean generation intervals ( $\pm$  standard error), in years, for the four pathways parent–offspring computed using the whole pedigree of the Gochu Asturcelta pig breed

Pathway	<i>n</i>	Years
Boar–son	100	2.0 ( $\pm$ 0.09)
Boar–daughter	264	1.7 ( $\pm$ 0.05)
Sow–son	100	1.8 ( $\pm$ 0.09)
Sow–daughter	264	1.7 ( $\pm$ 0.06)
Average	728	1.8 ( $\pm$ 0.03)

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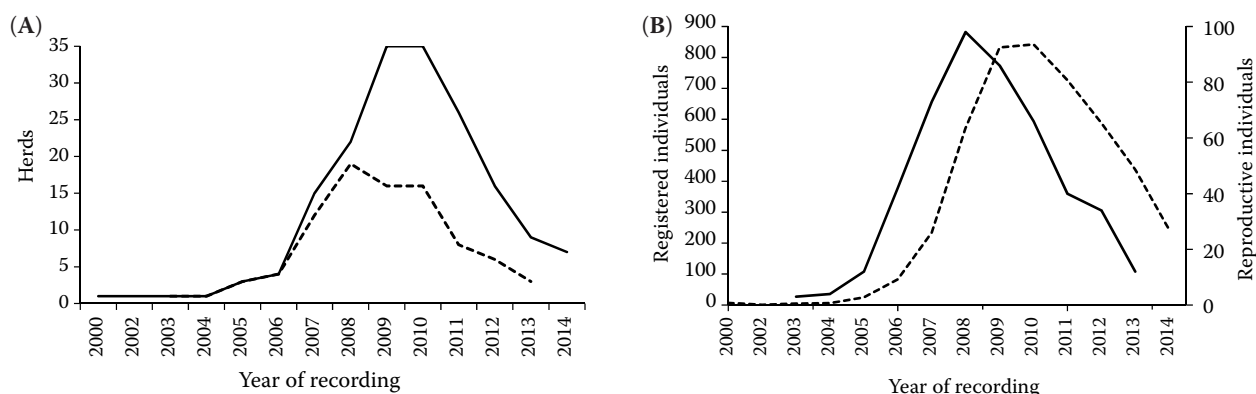


Figure 1. Number of registrations in the Gochu Asturcelta herdbook by years in the analyzed dataset

total (dashed line) and active (herds giving offspring; solid line) number of herds in the herdbook (A), total (dashed line) and the number of individuals giving offspring (solid line) registered in the herdbook (B)

als giving offspring to the herdbook during that year of recording.

The average generation interval computed was 1.8 ( $\pm 0.03$ ) years. The period for selection of females for reproduction was always shorter (1.7 years) than the period for selection of new boars ( $2.0 \pm 0.09$  years for the boar–son pathway) (Table 1).

**Parameters characterizing genetic diversity.** Table 2 gives parameters characterizing genetic diversity in the two reference populations defined and in the whole pedigree of the Gochu Asturcelta pig breed. The average number of equivalents to

complete generations was higher than 5 for the two reference populations fitted and  $4.3 \pm 0.9$  for the whole pedigree.

The total number of founders identified was 4 and 5 for the two reference populations defined, PopO and PopR, respectively, and 6 for the whole pedigree. Despite this extremely low number of founders, variability was significantly preserved. The values of  $f_e$  and  $f_a$  were basically the same for the two reference populations and for the whole pedigree. Surprisingly, the ratio of  $f_a$  to  $f_e$  is higher than 1 for the whole pedigree. This unexpected

Table 2. Parameters characterizing the probability of gene origin of the animals included in the two reference populations defined: animals born and registered in 2013 and 2014 (PopO) and animals born in 2012 and 2013 and kept for reproduction (PopR). The same parameters are given for the whole pedigree defined as all the individuals with both parents known (default definition of a reference population implemented in the program ENDOG)

	PopO	PopR	Whole pedigree
Total number of animals in the population	232	18	3149
Total number of founders	4	5	6
Effective number of founders ( $f_e$ )	3.50	3.48	3.47
Effective number of ancestors ( $f_a$ )	3.48	3.47	4.19
Founder genomes equivalents ( $f_g$ )	1.95	1.97	2.23
Effective number of non-founders ( $nf_e$ )	4.40	4.54	6.24
Ancestors explaining 100% of genetic variability	4	5	10
Ancestors explaining 50% of genetic variability	2	2	2
Inbreeding ( $F$ )	$0.289 \pm 0.085$	$0.249 \pm 0.070$	$0.230 \pm 0.078$
Average relatedness (AR)	$0.448 \pm 0.018$	$0.450 \pm 0.022$	$0.447 \pm 0.038$
Individual increase in inbreeding ( $\Delta F_i$ )	$0.067 \pm 0.021$	$0.067 \pm 0.021$	$0.079 \pm 0.034$
Equivalent to complete generations ( $t$ )	$5.4 \pm 0.5$	$5.2 \pm 0.4$	$4.3 \pm 0.9$
$N_e F_i$	$6.45 \pm 0.91$	$7.44 \pm 0.87$	$6.29 \pm 1.08$
$N_e C_{ij}$	$9.15 \pm 0.54$	$9.91 \pm 0.41$	$7.09 \pm 0.52$
$F_{IS}$	0.044	–0.007	0.008

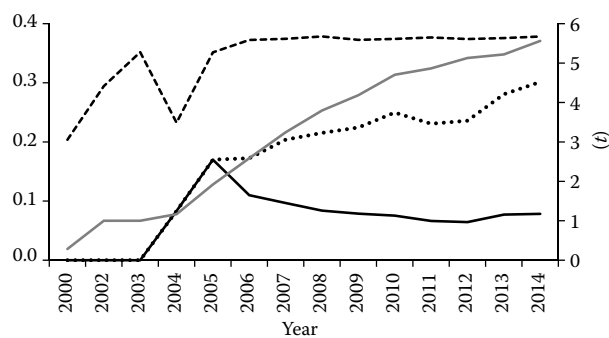


Figure 2. Average inbreeding values (dotted line), mean individual increase in inbreeding (solid black line), mean average relatedness (AR; dashed line), and average equivalent to complete generations (t; solid grey line) in the Gochu Asturcelta pig breed herdbook by year of birth of the individuals

result illustrated that the method of Boichard et al. (1997) was not able to identify an abusive use of some individuals for reproduction that would have led to a bottleneck in the population. Alternatively, losses of diversity caused by drift were assessed using the effective number of non-founders, which could be interpreted in a similar manner as  $f_a$  (Caballero and Toro 2000). Parameter

$nf_e$  was higher than  $f_a$ , and even higher than the actual number of founders, except for PopR, for each analyzed population (Table 2). In any case, drift caused noticeable losses of genetic diversity:  $f_g/f_e$  ratio was around 0.6 for the two reference populations (0.64 for the whole pedigree).

Figure 2 shows the variation of inbreeding, individual increase in inbreeding, average relatedness, and number of equivalents to discrete generations by year of birth of the individuals. As expected, both  $t$  and  $F$  steadily increased with years to reach values of  $0.30 \pm 0.09$  and  $5.5 \pm 0.59$ , respectively, in 2014. However, after an initial sudden increase (0.17 in 2005)  $\Delta F_i$  kept values around 0.07 after 2009. This is consistent with the variation assessed for AR which has kept steady on 0.37–0.38 since 2006. Note that AR is roughly two-fold coancestry. Table 2 gives the mean  $F$ , AR, and  $\Delta F_i$  values for the assessed populations. PopO had the highest mean  $F$  ( $0.289 \pm 0.085$ ) while this parameter took a value of  $0.230 \pm 0.078$  for the whole pedigree. Mean AR values computed were quite similar whatever the reference population considered ( $0.447 \pm 0.038$  for the whole pedigree). Mean  $\Delta F_i$

Table 3. Description of the founders<sup>1</sup> and ancestors<sup>2</sup> identified for each reference population defined in the Gochu Asturcelta pig breed herdbook

Founders/ancestors	Father	Mother	Sex	Year	Average relatedness (%)			Explained variability <sup>3</sup> (%)		
					whole pedigree	PopO	PopR	whole pedigree	PopO	PopR
0004 Sieru	9000 L'Utiru	9001 La Preñá	male	2003				36.28		
0001 L. láscaras	9000 L'Utiru	9001 La Preñá	female	2003				24.10		
0008 Kéndanu	9002 Machu	9006 Fema 2	male	2004				14.48	19.05	16.49
0009 Manteiga	9002 Machu	9006 Fema 2	female	2004				14.05	12.15	11.72
0005 Pola	9000 L'Utiru	9001 La Preñá	female	2003				8.83		
0010 Leonor	0006 Salva	9003 Fema 1	female	2005				1.97		
9000 L'Utiru			male	2000	35.19	34.40	35.16	0.12	34.40	35.16
9001 La Preñá			female	2000	35.19	34.40	35.16	0.12	34.40	35.16
9005 Tixu			male	2004	0.06			0.03		
9003 Fema 1			female	2000	1.03		1.48	0.02		1.48
9002 Machu <sup>4</sup>			male	2000	14.27	15.60	14.11			
9006 Fema 2 <sup>4</sup>			female	2000	14.27	15.60	14.11			

PopO = reference population including the offspring born in 2013 and 2014, PopR = reference population including the individuals born in 2012 and 2013 and kept for reproduction

<sup>1</sup>founders are those with non-zero average relatedness coefficients

<sup>2</sup>ancestors are those with non-zero explained variability coefficients

<sup>3</sup>computed using the method of Boichard et al. (1997) (see Material and Methods section)

<sup>4</sup>non-ancestor founder



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was the same for PopO and PopR ( $0.067 \pm 0.021$ ) but higher for the whole pedigree ( $0.079 \pm 0.034$ ).

The estimates of  $N_e$  given in Table 2 varied with both the methodology used and the dataset assessed. Estimates were always higher when the coancestry-based method was used. Estimates of  $N_e F_i$  varied from  $6.29 \pm 1.08$  (whole pedigree) to  $7.44 \pm 0.87$  (PopR). In turn, estimates of  $N_e C_{ij}$  varied from  $7.09 \pm 0.52$  (whole pedigree) to  $9.91 \pm 0.41$  (PopR).

Even though  $F_{IS}$  took a negative value for PopR, this parameter was roughly 0 for PopR and for the whole pedigree. However,  $F_{IS}$  was positive and high (0.044) for PopO.

Table 3 gives a description of the genetic variability explained by the founders and ancestors identified for each reference population. One of the actual founders of the breed (Tixu) had no viable grandsons and, therefore, its contribution to the breed was negligible ( $AR = 0.06\%$ ) and nonexistent in the present reference populations (PopO and PopR). One female founder (Fema 1) had a very poor contribution to the breed but it still remained represent in PopR (contribution of 1.48%). The other four founders formed two families (L'Utiru  $\times$  La Preñá and Machu  $\times$  Fema 2) which explained most of the genetic variability of the breed. In the whole pedigree, analyses could not find a clear bottleneck after the foundation of the breeding programme and both the founders L'Utiru and La Preñá and their descendants were identified as ancestors. Moreover, L'Utiru and La Preñá were identified as the main ancestors of the breed for PopO and PopR together with the offspring of Machu  $\times$  Fema 2.

## DISCUSSION

**Demographic analyses.** Average generation interval computed for Gochu Asturcelta (1.8 years) is consistently the same as the others previously reported in the literature for non-endangered pig breeds. Melka and Schenkel (2010) reported generation intervals of 1.6 and 1.7 years for Canadian Duroc and Landrace, respectively; Welsh et al. (2010), analyzing the pedigree of five United States pig breeds, reported generations intervals ranging from 1.7 (Berkshire) to 2.2 (Yorkshire) years; Tang et al. (2013), in three cosmopolitan pig breeds imported into China, reported generation intervals ranging between 1.8 and 1.9 years.

In contrast, the computed generation interval is lower than that of 2.45 years (ranging from 1.92 to 3.17 years for different cohorts) reported for the 'El Dehesón del Encinar' herd of Iberian pig (Toro et al. 2000) kept closed since its foundation in 1944. The very particular genetic scenario of the Gochu Asturcelta pig breed, characterized by a very low founder population size, could have led to an enlargement of the generation interval to avoid inbreeding accumulation. However, the interest in supplying with reproductive individuals to an increasing number of farmers has caused that the population dynamics of the breed is closer to that of a commercial pig breed than to that of a pig population under a preservation programme. In any case, this commercial-like breeding pattern topped out after 2010 when the possibilities of recruitment of new farmers decreased (Figure 1).

**Genetic scenario.** In any case, the general scenario of the Gochu Asturcelta pig breed can hardly be compared with others in the literature. Indeed, pedigree depth is substantially lower than the depths reported for the well-established commercial pig breeds of Canada and the United States (Melka and Schenkel 2010; Welsh et al. 2010) which included, in general, more than 11 complete generation equivalents. Nevertheless, the mean inbreeding in the breed is up to seven-fold higher than that reported for these American pig populations, ranging from 0.032 to 0.078. Furthermore, most estimates of effective population sizes available in the literature (usually ranging from  $N_e = 72$  to  $N_e = 125$ ) have been obtained in pig breeds with founder populations including hundreds or thousands individuals and rates of inbreeding per generation lower than 1% (Melka and Schenkel 2010; Welsh et al. 2010; Tang et al. 2013). Melka and Schenkel (2010) reported  $N_e = 14$  for the Canadian Hampshire pig which had a founder population size of 257 individuals and a rate of inbreeding of 0.036. Toro et al. (2000) reported an average population size of 13.8 for the Iberian pig herd of 'El Dehesón del Encinar' corresponding to a rate of inbreeding of 0.022. The  $N_e$  reported by Toro et al. (2000), for a population derived from 20 males and 82 females which initiated a closed breeding programme in 1944 (Silio and Rodriganez 2013), is probably overestimated due to the fact that it was computed using family variances. In a scenario of overlapping generations, computation of  $N_e$  based on family variances ignores several causes of vari-

ation of the parameter, namely mating between relatives or different representation of founders (Gutierrez et al. 2008). To illustrate this point we have computed  $N_e$  based on family variances for each biannual period (mimicking the average generation interval) from 2007 to 2010 using the program ENDOG. The values obtained varied from  $N_e = 21.3$  (period 2009–2010) to  $N_e = 34.5$  (period 2007–2008) which are clear overestimates of the effective sizes computed for the reference populations PopR and PopO (Table 2).

The main estimates of  $N_e$  given in the current analysis for the Gochu Asturcelta pig ( $N_e F_i$ ), whatever the reference population considered, roughly resembled the actual number of founders of the breed. Considering the high mean  $F$  of the pedigree analyzed, lower  $N_e$  values would be likely. However, after a sudden increase of  $F$  during the first stages of the breeding programme due to wrong breeding practices including full-sib matings (Menendez et al. 2015), the breeding policy avoided matings between close relatives as much as possible (Figure 2). This led to minimizing the increase in AR and even to a decrease in mean  $\Delta F_i$  in the last few years of pedigree recording. In such scenario, computation of  $N_e$  via regression of  $F$  on  $t$  (Gutierrez et al. 2003) gave unrealistic estimates including negative  $N_e$  values for PopO (data not shown). The current estimates of  $N_e F_i$  are not affected by mating policy or bottlenecks caused by poor use of reproducing individuals (Cervantes et al. 2008). Note that the genealogical differentiation of the two reference populations fitted (PopR and PopO) is negligible ( $F_{ST} = 0.0033$ ) and, therefore, it is not likely that the scenario described for PopO is affected by a very recent change in mating policy.

**Insights for conservation.** Whatever the  $N_e$  threshold chosen to consider a livestock breed to be at risk (Leroy et al. 2013), the Gochu Asturcelta pig breed is extremely endangered. Since one of the founders of the breed is not represented in the present population, the maximum number of founders available for the reference populations would be 5. These five founders are still genetically represented in PopR. This fact enables us to consider that the breeding policy of the ACGA has been successful. Despite the value of  $F_{IS}$  was high and positive for PopO, this parameter was basically zero in the whole pedigree and even negative for PopR. Parameter  $F_{IS}$  is equivalent to the parameter alpha proposed by Caballero and

Toro (2000) and informs on the avoidance (or not) of matings between relatives, thus characterizing the breeding policy. In the whole pedigree and, particularly, for the selection of the parents for the next generation (PopR), breeders follow strict avoidance of matings between close relatives. Most of the individuals forming PopO will supply local market demands for quality pork meat.

Related to that, one of the most striking features of the current analysis is that  $f_a > f_e$  for the whole pedigree. Even though that did not happen for the smaller reference populations (PopO and PopR), the method of Boichard et al. (1997) failed to identify bottlenecks due to an abusive use of reproductive individuals in the whole pedigree. This was confirmed by the parameter  $nf_e$  which would have a similar interpretation than  $f_a$  (Caballero and Toro 2000). While  $f_a$  is calculated using an approximate method that “creates” new ancestors via identifying bottlenecks in which genealogical paths converge, to further compute the marginal contribution of an ancestor (Boichard et al. 1997), parameter  $nf_e$  is computed directly on data in an exact way (Caballero and Toro 2000). The direct descendants of the founders of the pedigree were quickly included in the multiplier herd of SERIDA and its reproductive lifespan was prolonged as much as possible including crosses with their descendants of the first and second generations. For the whole pedigree, genealogical paths traced back to the base populations do not stop in the F1 individuals but still can be traced back to the founders to some extent (Table 3). Furthermore, parameter  $nf_e$  shows that the random losses of genes during the segregations, after the breeding programme was initiated, were minimized as much as possible.

Furthermore, comparison of estimates of  $N_e F_i$  and  $N_e C_{ij}$  can give insights for the conservation programme of the Gochu Asturcelta pig breed. Since  $N_e F_i$  and  $N_e C_{ij}$  should coincide under random mating, a ratio  $(\overline{N_e} - \overline{C_{ij}})/(\overline{N_e} F_i)$  higher than 1 would inform of some degree of population subdivision (Cervantes et al. 2011a, b). Consistently with the negative value of  $F_{IS}$ , this ratio is particularly high for PopR (1.23). Probably, this is due to a relatively high genetic representation of the founder 9003 Fema 1 in PopR (Figure 3). The genetic background of the breed is basically formed by two families (L’Utiru  $\times$  La Preña and Machu  $\times$  Fema 2). However, the genetic variability explained by the founder 9003 Fema 1 as an

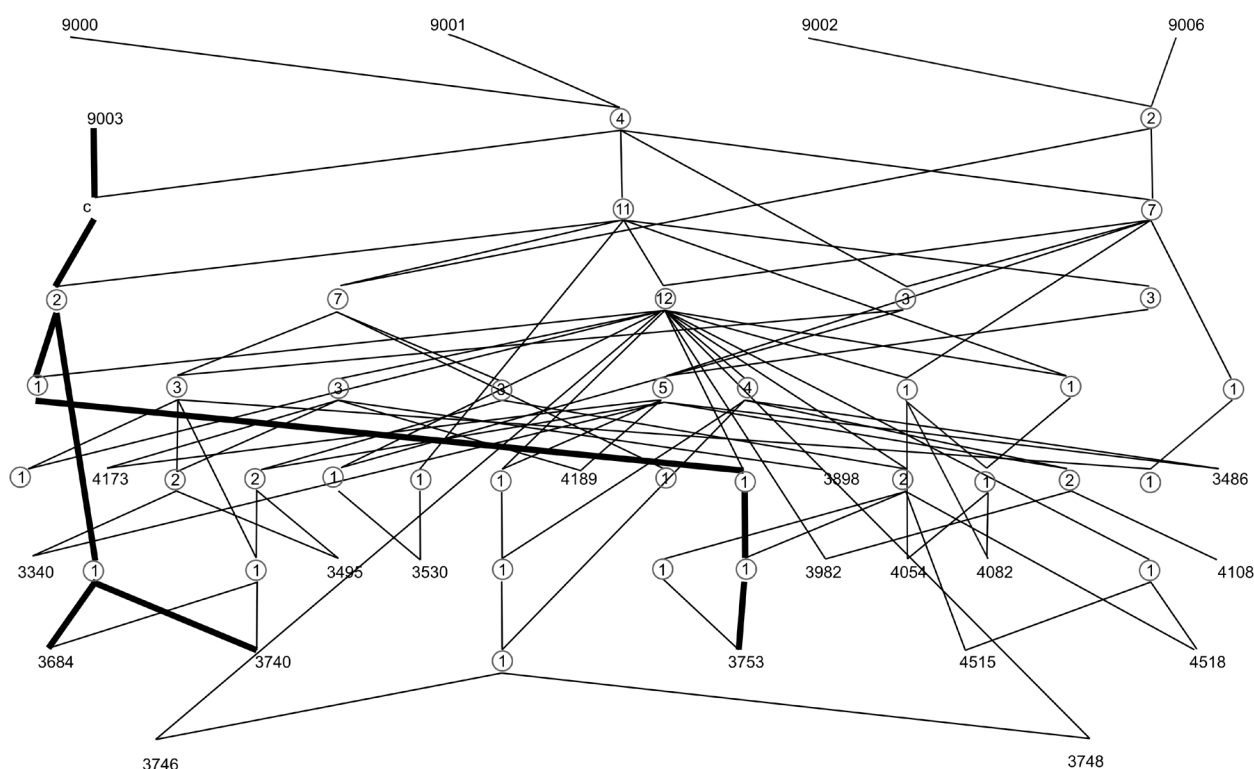


Figure 3. Summary of the pedigree of 18 individuals born in 2012 and 2013 and kept for reproduction used to form the reference population PopR (see Material and Methods section). ID of founders: 9000–9006, ID of PopR individuals: 3340–4518. Paths have been simplified as much as possible: circles in the nodes include the number of reproductive individuals in the pedigree which are descendants of the same cross. Paths involving founder 9003 are in thick lines

ancestor in the most recent reproductive population (PopR) is considerably higher than in the whole pedigree (1.48% vs 0.02%). When the genetic representation of some lines of founders vastly outweighs others in the present population of an endangered breed it has been suggested to unbalance the genetic contributions of specific individuals to equalize the genetic representation of the founders and lines in the population (Ballou and Lacy 1995). In the endangered Asturian livestock breeds parameter AR is routinely used to monitor pedigrees (Goyache et al. 2003; Royo et al. 2007; Alvarez et al. 2008). Descendants of under-represented founders are identified and offspring in the further few breeding seasons is unbalanced as much as possible towards those animals to maintain the initial genetic variability and to control average AR values in the new stock. In the Gochu Asturcelta pig breed, new individuals kept for reproduction, males if possible, must be intentionally selected from descendants of the founder 9003 Fema 1 to delay, to some extent, losses of genetic diversity.

## CONCLUSION

The current analysis illustrates that strict mating policies can be useful to maintain genetic variability in extremely small livestock outbred populations. Even if some founder lines contribute only slightly to the population, the avoidance of population bottlenecks after the implementation of a recovery programme for a breed is possible. Furthermore, continuous monitoring of pedigrees has proved to be useful to evaluate the effects of the mating policy and to identify individuals with low genetic representation in the population to allow further balance of genetic representation of founders in the next generation.

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