ASSESSMENT OF GENETIC RISK AND LONG-TERM VIABILITY IN THREE SPECIALIZED PALAS SHEEP BREEDS

O.-C. Preșa (Dordescu)[1,2], Z. C. Zamfir[1*], A. G. Vartic[1,2], L. Marinică[2,3], A. Enciu[1], M. Stanciu[1], F. Leica[1], L. A. Liță[1], Ș. Creangă[2]

[1]Research and Development Institute for Sheep and Goat Breeding Palas-Constanța, 248 I.C. Brătianu Boulevard, Constanța, Romania
[2]Faculty of Food and Animal Sciences, Iași University of Life Sciences, No. 8, Mihail Sadoveanu Alley, Iași, Romania
[3]Faculty of Natural Sciences and Agricultural Sciences, Ovidius University of Constanța, Constanța, Romania

Abstract

This study characterized genetic risk and long-term viability in the Palas Milk Breed, Palas Meat Breed, and Prolific Palas Breed populations maintained at the Research and Development Institute for Sheep and Goat Breeding Palas-Constanța. The indicators analyzed were effective population size (Ne), effective number of breeding males (Nm), increase in inbreeding per generation (ΔF), generation interval (L), annual inbreeding rate (ΔFan), and estimated 50-year accumulation of inbreeding (ΔF50). The Palas Milk Breed population had Ne = 60.30, Nm = 17.12, ΔF = 0.73% and ΔFan = 0.19%/year; the Palas Meat Breed population had Ne = 93.11, Nm = 15.63, ΔF = 1.04%, and ΔFan = 0.29%/year and the Prolific Palas Breed population had Ne = 87.72, Nm = 21.93, ΔF = 0.57% and ΔFan = 0.13%/year. The annual inbreeding rates remained below 0.5%/year. The findings support the continued use of rotational mating, balanced contributions from sire families, and periodic pedigree monitoring in each population.

Key words: *effective population size; inbreeding; genetic conservation; sheep; Palas*

INTRODUCTION

The conservation of animal genetic resources is a major component of sustainable livestock production because within-breed genetic diversity supports adaptive capacity, resilience, and the long-term response to selection. In small populations, genetic drift and the accumulation of inbreeding may progressively reduce heterozygosity and increase the probability of losing rare alleles. Effective population size (Ne), rather than census population size alone, is therefore a central parameter in the management of conserved populations because it reflects the number and balance of breeding animals that effectively contribute genes to subsequent generations [1,2].

Recent studies of European sheep populations have demonstrated that demographic and pedigree-based indicators remain valuable tools for monitoring genetic diversity. Analyses of German sheep breeds identified substantial variation in Ne and inbreeding rates among populations, confirming the value of periodic pedigree monitoring [3]. Genomic studies of local breeds from Portugal, Croatia, and Belgium further emphasize that conservation decisions should integrate population size, inbreeding, relatedness, and the contribution of male breeders

*Corresponding author: zoia.zamfir@icdcocpalas.ro
The manuscript was received: 7.10.2026
Accepted for publication: 10.11.2026

Article licensed under a Creative Commons Attribution-NonCommercial-ShareAlike 4.0 International License (http://creativecommons.org/licenses/by-nc-sa/4.0/)

[4,5,6]. More recent research highlights the importance of genetically informed mating strategies for small Balkan sheep populations [7] and confirms, at the genomic level, that controlling inbreeding is essential for preserving long-term breeding potential [8].

The three specialized Palas sheep breeds are maintained as closed or controlled populations under conservation and breeding programs at the Research and Development Institute for Sheep and Goat Breeding Palas-Constanța. Because previous publications have addressed breed development and genetic history, the present study does not re-examine breed origins or founder contributions. It focuses exclusively on current population indicators that are directly relevant to genetic risk assessment and conservation management.

The objective of this study was to assess the Palas Milk Breed, Palas Meat Breed, and Prolific Palas Breed populations using effective population size (Ne), effective number of breeding males (Nm), increase in inbreeding per generation (ΔF), generation interval (L), annual inbreeding rate (ΔFan), and estimated 50-year accumulation of inbreeding (ΔF50), and to define genetic management priorities for each breed.

MATERIAL AND METHOD

The analysis included the Palas Milk Breed, Palas Meat Breed and Prolific Palas Breed populations maintained at the Research and Development Institute for Sheep and Goat Breeding Palas-Constanța. Population records and pedigree information from the conserved flocks were used. The reported census sizes were 520 animals for Palas Milk Breed, 570 for Palas Meat Breed, and 410 for Prolific Palas Breed. The analysis did not include a historical reconstruction of breed development.

Effective population size (Ne) was evaluated from the numbers of breeding males (Nm) and females (Nf) using the standard sex-ratio equation $Ne = \frac{4Nm \times Nf}{Nm+Nf}$, which is frequently applied in the management of animal genetic resources [1,2]. The effective number of breeding males was estimated from the reproductive structure and the contribution of males to the next generation within each population. The populations are organized into sire families and managed through rotational mating among unrelated or weakly related families.

The increase in inbreeding per generation (ΔF, %) was evaluated from the pedigree and reproductive data available for each population. The generation interval (L, years) was calculated from the corresponding pedigree information. The annual inbreeding rate was calculated as $\Delta Fan = \frac{\Delta F}{L}$ and long-term accumulation was expressed as $\Delta F50 = 50 \times \Delta Fan$. These projections describe the expected accumulation under the current reproductive management conditions and should be interpreted as monitoring indicators rather than fixed biological outcomes.

For descriptive interpretation, annual inbreeding rates below 0.5% were considered low and compatible with controlled conservation management, whereas increasing values were considered indicators requiring progressively closer monitoring. Because the study examined complete conserved populations rather than random experimental samples, the results are presented descriptively.

RESULTS

The Palas Milk Breed population comprised 520 animals, with Ne = 60.30 and Nm = 17.12. The Palas Meat Breed population comprised 570 animals, with Ne = 93.11 and Nm = 15.63. The Prolific Palas Breed population comprised 410 animals, with Ne = 87.72 and Nm = 21.93. These values are presented in Figure 1 as descriptive parameters of the reproductive structure of each population.

Article licensed under a Creative Commons Attribution-NonCommercial-ShareAlike 4.0 International License (http://creativecommons.org/licenses/by-nc-sa/4.0/)

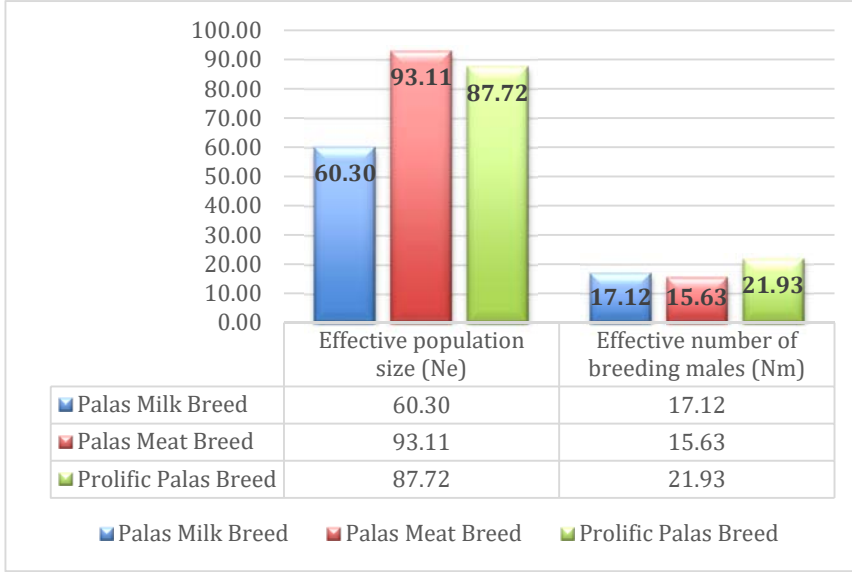


Fig. 1 Effective population size and breeding males

The estimated inbreeding parameters and generation intervals for the three Palas sheep breeds are presented in Table 1. Palas Milk Breed showed an increase in inbreeding (ΔF) of 0.73% per generation, with a generation interval of 3.84 years, corresponding to an annual inbreeding rate (ΔFan) of 0.19%/year. In Palas Meat Breed, ΔF reached 1.04% per generation, with a generation interval of 3.65 years and an annual inbreeding rate of 0.29%/year. The lowest values were recorded in Prolific Palas Breed, with ΔF of 0.57% per generation, a generation interval of 4.49 years, and an annual inbreeding rate of 0.13%/year.

Table 1 Inbreeding parameters and generation interval in the three Palas sheep breeds

Breed	ΔF (%/generation)	Generation interval, L (years)
Palas Milk Breed	0.73	3.84
Palas Meat Breed	1.04	3.65
Prolific Palas Breed	0.57	4.49

The annual inbreeding rates and the projected accumulation of inbreeding over a 50-year period are illustrated in Figure 2. Assuming that the current breeding and mating management conditions remain unchanged, the linear projection indicated a 50-year accumulation of 9.50% for Palas Milk Breed, 14.25% for Palas Meat Breed and 6.35% for Prolific Palas Breed. These projections should not be interpreted as fixed future outcomes, but rather as monitoring indicators describing the expected trajectory of inbreeding under the continuation of current management practices.

Article licensed under a Creative Commons Attribution-NonCommercial-ShareAlike 4.0 International License (http://creativecommons.org/licenses/by-nc-sa/4.0/)

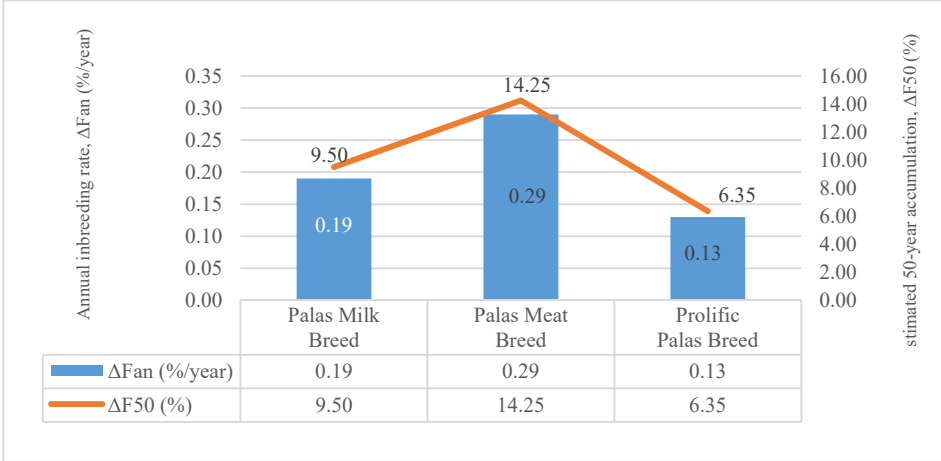


Fig. 2 Annual inbreeding rate (ΔFan) and estimated 50-year accumulation of inbreeding (ΔF50)

The conservation profile of the three Palas sheep breeds, together with the main management priorities derived from the estimated demographic and inbreeding parameters, is summarized in Table 2. The results indicate differences among breeds in the parameters that require closer monitoring. Palas Milk Breed showed a relatively low annual inbreeding rate, although its effective population size suggests the need for continued periodic surveillance. Palas Meat Breed had the highest effective population size, but also showed the highest annual inbreeding rate and the highest projected 50-year accumulation of inbreeding, indicating the importance of controlling the use of breeding males and maintaining a balanced family contribution. Prolific Palas Breed showed the lowest annual inbreeding rate and the lowest projected long-term accumulation, while preservation of the current breeding base and family structure remains important for maintaining genetic diversity.

Table 2 Conservation profile and management priorities for each breed

Breed	Main favorable indicator	Main aspect requiring attention
Palas Milk Breed	Annual inbreeding rate of 0.19%/year	Ne = 60.30; periodic monitoring required
Palas Meat Breed	Ne = 93.11	Nm = 15.63; ΔFan = 0.29%/year; ΔF50 = 14.25%
Prolific Palas Breed	Nm = 21.93; ΔFan = 0.13%/year; ΔF50 = 6.35%	Maintain the breeding base and family structure

The recommended conservation and breeding management priorities for the three Palas sheep breeds are presented in Figure 3. The proposed priorities were established by integrating the effective population size, number of breeding males, annual inbreeding rate, and projected long-term accumulation of inbreeding. Particular emphasis should be placed on maintaining a balanced contribution of breeding animals, avoiding excessive use of a limited number of sires, and periodically monitoring inbreeding trends and family structure.

Article licensed under a Creative Commons Attribution-NonCommercial-ShareAlike 4.0 International License (http://creativecommons.org/licenses/by-nc-sa/4.0/)

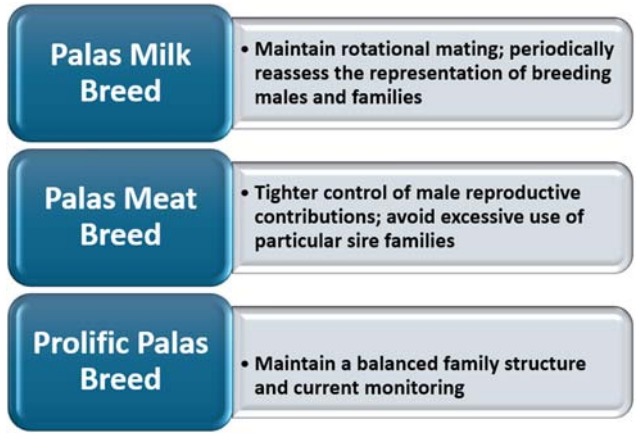


Fig. 3 Recommended conservation and breeding management priorities for the three Palas sheep breeds

DISCUSSIONS

The present findings demonstrate why census population size should not be used alone to assess genetic security. In Palas Meat Breed, a census size of 570 animals and Ne = 93.11 coexist with Nm = 15.63 and ΔF = 1.04% per generation. This association is biologically plausible because effective population size and future inbreeding are influenced by the sex ratio, variation in family size, and unequal reproductive contributions. FAO recommendations emphasize that using a limited number of males can reduce genetic diversity and accelerate genetic drift even when census population size is relatively large [1,2].

The observed Ne values, ranging from 60.30 to 93.11, indicate the need for continued active genetic management. In a large pedigree analysis of 35 German sheep breeds, Justinski et al. [3] reported realized Ne estimates within a similar range and showed that genetic diversity may continue to decline even in breeds with apparently adequate population sizes. The Palas populations should therefore be managed as genetic resources in which balanced family contributions remain essential.

Comparable types of indicators have also been reported for other sheep populations. Justinski et al. [3] reported a mean realized Ne of 91.4 and a mean generation interval of 3.83 years across 35 German breeds analyzed from pedigree data. In six Irish breeds, Rafter et al. [9] estimated Ne values ranging from 116.0 to 314.8 together with the effective numbers of founders, ancestors, and founder genomes. In French dairy sheep breeds, Rodríguez-Ramilo et al. [10] assessed Ne using pedigree, SNP-by-SNP, and ROH approaches. These findings provide methodological benchmarks, but direct numerical equivalence is not warranted when estimation methods and reference periods differ.

For Prolific Palas Breed, Nm = 21.93, ΔF = 0.57%, and ΔFan = 0.13%/year indicate a reproductive structure compatible with maintaining the contribution of sire families under the current program. Studies of European local breeds likewise show that limiting inbreeding and maintaining effective population size depend on avoiding the excessive use of a restricted number of male breeders [4,5,6,7].For Palas Meat Breed, Nm = 15.63 and ΔF = 1.04% per generation support close monitoring of sire contributions. The annual inbreeding rate was 0.29%/year, and the estimated 50-year accumulation was 14.25%. These values do not indicate an immediate loss of

Article licensed under a Creative Commons Attribution-NonCommercial-ShareAlike 4.0 International License (http://creativecommons.org/licenses/by-nc-sa/4.0/)

viability, but they support measures to prevent the disproportionate representation of particular paternal families. Pedigree and genomic analyses of endangered Belgian sheep populations have similarly demonstrated the need for continuous monitoring even where structured breeding programs are in place [6].

The study also supports the continued use of rotational mating among unrelated or weakly related sire families. Such strategies are particularly valuable when the number of breeding males is limited because they can restrict the rate of accumulation of close relationships. Genetically informed conservation studies of Balkan sheep populations similarly emphasize the importance of avoiding matings between closely related animals and maintaining multiple paternal lines [7].

A limitation of this study is that the analysis is based on demographic and pedigree-derived parameters rather than estimates obtained from genome-wide SNP markers or whole-genome sequencing. Genomic methods, particularly runs of homozygosity (ROH) analysis and genomic estimates of Ne, can reveal recent autozygosity and cryptic relationships that are not fully captured by pedigree data [4,5,8]. A logical next step would therefore be to integrate the existing pedigree monitoring system with genomic estimates of inbreeding and ROH analysis. This approach would enable independent validation of the current conservation programs without repeating previous investigations of breed genetic history.

CONCLUSIONS

The indicators analyzed describe the conservation characteristics of each Palas population. For Palas Milk Breed, rotational mating should be maintained and family representation periodically reassessed. For Palas Meat Breed, the contribution of male breeders should be monitored and repeated use of the same paternal lines avoided. For Prolific Palas Breed, family structure should be preserved and pedigree monitoring continued. Ne should be interpreted together with Nm and the inbreeding rate, while future genomic analyses may complement the pedigree-based assessment.

ACKNOWLEDGMENTS

This research was conducted within project ADER 7.2.1, 'Research on maintaining biodiversity in sheep by assessing the risk status of specialized sheep breeds, conserving and using their gene pools to disseminate genetic progress in sheep farms', funded through the Sectoral Research and Development Plan of the Romanian Ministry of Agriculture and Rural Development.

REFERENCES

1. Rischkowsky, B; Pilling, D, Eds. *The State of the World's Animal Genetic Resources for Food and Agriculture*; Food and Agriculture Organization of the United Nations: Rome, Italy, **2007**.
2. Hodges, J, Ed. *The Management of Global Animal Genetic Resources*; FAO Animal Production and Health Paper No. 104; Food and Agriculture Organization of the United Nations: Rome, Italy, **1992**.
3. Justinski, C; Wilkens, J; Distl, O Genetic Diversity and Trends of Ancestral and New Inbreeding in German Sheep Breeds by Pedigree Data. *Animals* **2023**, *13*, 623. doi:10.3390/ani13040623.
4. Gaspar, D; Usié, A; Leão, C; Guimarães, S; Pires, A.E; Matos, C; Ramos, AM; Ginja, C Genome-Wide Assessment of the Population Structure and Genetic Diversity of Four Portuguese Native Sheep Breeds. *Front. Genet.* **2023**, *14*, 1109490. doi:10.3389/fgene.2023.1109490.
5. Ramljak, J; Špehar, M; Ceranac, D; Držaić, V; Pocrnić, I; Barać, D; Mioč, B; Širić, I; Barać, Z; Ivanković, A; Kasap, A Genomic Characterization of Local Croatian Sheep Breeds—Effective Population Size, Inbreeding & Signatures of Selection. *Animals* **2024**, *14*, 1928. doi:10.3390/ani14131928.
6. Meyermans, R; Gorssen, W; Aerts, N; Hooyberghs, K; Chakkingal Bhaskaran, B;

Article licensed under a Creative Commons Attribution-NonCommercial-ShareAlike 4.0 International License (http://creativecommons.org/licenses/by-nc-sa/4.0/)

Chapard, L; Buys, N; Janssens, S Genomic Characterisation and Diversity Assessment of Eight Endangered Belgian Sheep Breeds. *Animal* **2024**, *18*, 101315. doi:10.1016/j.animal.2024.101315.
7. Ćosić, I; Stojiljković, KZ; Pihler, I.; Cekić, B.; Ružić-Muslić, D.; Delić, N.; Aleksić, J.M. Towards Genetically Informed Conservation of the Bardoka and Karakachan Sheep Breeds Autochthonous to Serbia. *Animals* **2025**, *15*, 1193. doi:10.3390/ani15091193.
8. Ma, R; Liu, J; Ma, X; Yang, J Genome-Wide Runs of Homozygosity Reveal Inbreeding Levels and Trait-Associated Candidate Genes in Diverse Sheep Breeds. *Genes* **2025**, *16*, 316. doi:10.3390/genes16030316.
9. Rafter, P; McHugh, N; Pabiou, T; Berry, DP Inbreeding Trends and Genetic Diversity in Purebred Sheep Populations. *Animal* **2022**, *16*, 100604. doi:10.1016/j.animal.2022.100604.
10. Rodríguez-Ramilo, ST; Elsen, JM; Legarra, A Inbreeding and Effective Population Size in French Dairy Sheep: Comparison between Genomic and Pedigree Estimates. *J. Dairy Sci.* **2019**, *102*, 4227–4237. doi:10.3168/jds.2018-15405.

Article licensed under a Creative Commons Attribution-NonCommercial-ShareAlike 4.0 International License (http://creativecommons.org/licenses/by-nc-sa/4.0/)