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DAGENE
International Association for the Conservation
of Animal Breeds in the Danube Region
1078 Budapest, István street 2.
Hungary



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This journal, founded 2016 is the official publication medium for the yearly activity of DAGENE members. The profile of the journal, reflecting the Association's mission, is to research, evaluate, present and document the conservation of endangered domestic animal breeds and their frontier areas in a scientific manner. The journal is mainly waiting for manuscripts on breed preservation in the Danube Valley from members of the Association, but also welcomes manuscripts from other dedicated authors and PhD students from other regions of the world, in English. Articles are peer-reviewed by experts in the preservation of biological diversity before publishing to ensure the article's quality. Manuscripts should be submitted to <http://ojs3.mtak.hu/index.php/dagr>.

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Comparison of two freezing techniques and two seminal extenders and their effect on post-thaw motility in the cryopreservation of goat sperm

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Abstract

The aim of this study was to compare the effect of two freezing techniques (programmable and manual) and two seminal extenders (Triladyl® and OptiXcell®) on the quality of cryopreserved goat sperm after thawing. The experiment involved ejaculates from goats of the Slovak White Shorthair Goat (n = 3) and Slovak Brown Shorthair Goat (n = 2), collected during the natural reproductive period of goats. After collection, the sperm were diluted, equilibrated for two hours at 4 °C and then frozen using either programmable or manual technique. After thawing, the progressive sperm motility was evaluated. The results showed that the cryopreservation process led to a decrease in sperm motility compared to fresh samples. However, the use of the OptiXcell® extender ensured significantly higher progressive sperm motility after thawing compared to the Triladyl® extender (65.38% vs. 40.06%; $p < 0.05$) for both freezing techniques. On the contrary, no statistically significant differences were found between programmable and manual freezing, indicating their comparable effectiveness (36.85% vs. 40.06% and 57.13% vs. 65.38%, respectively). Based on the results obtained, it can be concluded that the type of extender used has a more significant impact on sperm quality after thawing than the freezing technique itself. Manual freezing appears to be a suitable alternative to programmable freezing, especially under conditions with no access to automated equipment, which may have significant practical benefits for the preservation of goat genetic resources in gene banks.

Keywords: cryopreservation, goat, semen, Triladyl, Optixcell

Introduction

Goats are one of the most widespread animal species in the world and offer various uses and benefits (HANIFI et al., 2025). The White Shorthair goat is a dairy breed, developed in the first half of the 20th century by crossbreeding native local goats with imported Saanen breed goats from Switzerland and Germany. The goats are early maturing, hardy, highly fertile with good feed conversion ability. The Brown Shorthair goat breed was developed by crossbreeding native spotted and brown goats with imported Harz breed goats from Germany. Initially bred as hornless, today hornedness is permitted in both sexes. The black triangle behind the ears is a characteristic feature of the breed (CHRENEK et al., 2019).

Sperm cryopreservation plays a key role in the long-term ex-situ in vitro conservation of endangered breeds (RANJAN et al., 2021). It is an important tool for long-term preservation of genetic material, which allows the preservation and transmission of genetic traits through assisted reproduction (ARIYAN et al., 2025). This technique is also important for the breed conservation process and has paved the way for other reproductive biotechnologies. Despite significant progress in this field, frozen and thawed sperm have huge differences in fertility rates (RANJAN et al., 2021). One of the main challenges is the cryosensitivity of caprine sperm, as the lipid-rich membranes of caprine sperm are very susceptible to damage during freezing and thawing. The high lipid content of seminal plasma exacerbates this cryosensitivity, making it difficult to maintain sperm viability after thawing (KUMAR et al., 2025). Cryopreservation significantly impairs sperm quality in goats, resulting in reduced viability, motility, membrane integrity and increased DNA fragmentation. These impairments are primarily due to the excessive production of reactive oxygen species (ROS) during freezing and thawing, which overwhelms the natural antioxidant defences in sperm (TRAN et al., 2025). ROS, such as superoxide anion (O_2^-), hydroxyl radical ($-OH$) and hydrogen peroxide (H_2O_2), directly oxidize polyunsaturated fatty acids in sperm membranes, leading to lipid peroxidation and subsequent cellular dysfunction (SIKKA, 2004; LONE et al., 2018). The sperm membrane of goats is rich in unsaturated free fatty acids, which are susceptible to oxidative stress. Oxidative stress can alter the structure of the sperm head, leading to greater permeability of the cell membrane to fluids, resulting in lower sperm viability (MOHAMMED and OMAR, 2025). In addition, several factors such as breeds, seasons, husbandry practices and cryopreservation protocols are responsible for the poor quality of cryopreserved goat sperm (RANJAN et al., 2021).

Since the extent of cryodamage is strongly influenced by the composition of the preservation medium, the selection of an appropriate semen extender represents an important component of an effective cryopreservation protocol. Semen extenders provide an energy source, protect spermatozoa against cold shock and osmotic stress, and help maintain a suitable environment during cooling, freezing and thawing. A

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number of extenders and additives have therefore been developed and evaluated for both short- and long-term semen preservation (RANJAN et al., 2021).

The use of liquid sperm requires a diluent, because sperm stored at room temperature undergoes metabolism, which causes a decrease in sperm quality (HAFIDHA et al., 2025). A number of extenders and additives have been used and tested for short-term and long-term sperm storage to maintain sperm motility and viability during storage (ARI and YILDIZ, 2018). The most commonly used extenders for goat sperm are either egg yolk-based extenders or non-fat dry skimmed milk-based sperm extenders (RANJAN et al., 2021). Goats are unique among livestock because during sperm donation; they release specific lipase enzymes from their bulbourethral gland that interact with components of the sperm extensor (egg yolk lipids and skim milk triglycerides) to produce a compound that is toxic to sperm. Therefore, the lipase enzyme in the seminal plasma is called egg yolk coagulation enzyme (EYCE). The deleterious interaction between egg yolk and EYCE occurs only in sperm of goat origin but not in bulls, boars or rams (SIAS et al., 2005).

During cryopreservation, equilibration refers to the period after mixing sperm with a cryoprotectant (usually glycerol or other cryoprotectants) to allow the sperm to acclimate to subfreezing temperatures before freezing. Equilibration times of 1 to 2 hours at 4°C are usually recommended to allow the sperm to stabilize and facilitate the gradual diffusion of cryoprotectants into the sperm. If the equilibration time is too short, the sperm may not absorb the necessary amount of cryoprotectant resulting in poor protection during freezing and subsequent damage during thawing. On the other hand, excessive equilibration time of more than 3 hours may lead to overexposure to cryoprotectants, which may cause toxicity or membrane disruption resulting in lower sperm motility and fertility after thawing (KUMAR et al., 2025). Despite the progress made in the cryopreservation of caprine sperm, further improvements are needed to address issues such as oxidative stress, cryoinjury and environmental factors (KUMAR et al., 2025). The detailed molecular mechanism and extent of structural damage to sperm associated with cryopreservation have not been systematically investigated (RANJAN et al., 2021). Therefore, the aim of the present study was to compare the effects of two freezing techniques (manual and programmable freezing) and two commercial semen extenders (Triladyl® and OptiXcell®) on the progressive motility of cryopreserved goat sperm after thawing. In addition, the study aimed to determine whether manual freezing could provide post-thaw sperm motility comparable to that achieved using a programmable freezing system.

Material and methods

Animals

For these experiments, ejaculate samples from goats of the Slovak Brown Short-haired Goat ($n = 2$) and Slovak White Short-haired Goat ($n = 3$) breeds were used. During the experiments, the goats were kept separately from the goats in the breeding facility of the NPPC - Research Institute of Animal Production in Lužianky, at an altitude of approximately 154 m. a.s.l. Clinically healthy, sexually mature goats of the Slovak Brown Short-haired Goat and Slovak White Short-haired Goat breeds were involved in the experiments. The individuals were aged from 9 months to 3 years. The goats were fed meadow hay, alfalfa hay, oats (according to body condition score (BCS) and weight) and corn-barley meal (according to BCS and weight). Each goat had a vitamin-mineral supplement and water in its enclosure.

Collection and dilution of ejaculates

Sperm samples from goats were collected in 2025 during the natural reproductive period of goats (September – November) twice a week in the morning using an artificial vagina (Minitube, Germany) while jumping onto a fixed female. During the collection, the female was fixed in a standing position using simple physical sedation without the use of sedatives.

The sperm were processed immediately after collection at the laboratory of NPPC VÚŽV Nitra. From each goat, we diluted 10 μ l of fresh sample with saline solution (0.9% NaCl; Braun, Germany) as required. We diluted the goat sperm samples in the ratio of 1:40 (v/v) or 1:50 (v/v). For diluted samples, we evaluated the volume, concentration and progressive motility of freshly collected sperm. Subsequently, we exposed the collected goat sperm to the cryoprotective medium Triladyl® (Minitube, Germany) or OptiXcell® (IMV Technologies, France) in a ratio of 1:10 (v/v). Equilibration was carried out for 2 h at 4 °C according to BERČÍK et al. (2026).

Freezing techniques

Programmable freezing (PF)

After equilibration, the straws were placed in a pre-cooled automatic freezer (IceCube, Minitube, Germany). After closing the lid, the software started to regulate the flow of nitrogen vapor into the sample compartment according to the set freezing program (+4 °C to -10 °C for 120 s, -80 °C for 450 s, -120 °C for 100 s and -140 °C for 180 s). After completing the freezing cycle, the straws were immersed into liquid nitrogen (-196 °C) in a container (Liquid nitrogen container, KGW-Isotherm, Germany).

The straws were thawed in a water bath at 40 °C for 15 seconds, afterwards the samples were transferred into tubes for 5 min and then analysed.

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Manual freezing (MF)

We placed the filled straws 4 cm above the liquid nitrogen level on a floating stand in a freezing box (Freezing unit for 90 straws, Minitube, Germany) with liquid nitrogen for 15 minutes (reaching temperatures between -125 and -130 °C) and then slowly immersed them directly into liquid nitrogen (-196 °C).

To thaw, we immersed the straws into a bath for 15 seconds at 40 °C.

The two freezing procedures therefore differed primarily in the control and duration of the cooling phase. Programmable freezing involved a controlled, stepwise decrease in temperature from +4 °C to -140 °C over a total programmed period of 850 s (approximately 14.2 min) before immersion in liquid nitrogen (-196 °C). In comparison, manual freezing involved exposure of the straws to nitrogen vapour for 15 min at 4 cm above the liquid nitrogen surface, where temperatures of approximately -125 to -130 °C were reached, followed by direct immersion in liquid nitrogen. Thus, the overall duration of the freezing phase before immersion in liquid nitrogen was similar between the two techniques (approximately 14.2 vs. 15 min), although temperature reduction was precisely controlled during programmable freezing and passively achieved through nitrogen vapour exposure during manual freezing.

Statistical analyses

Cryopreservation experiments with both techniques were performed in triplicate. Differences between the studied groups were analysed using a one-way ANOVA. Tukey's post-hoc multiple comparison test was used to identify statistically significant differences between individual pairs of groups. The statistical significance level was set at $\alpha = 0.05$. For statistical processing, we used the GraphPad Prism program (version 9.0.0, GraphPad Software, USA).

Ethical approval

The experimental procedures were reviewed by the Ethics Committee of the National Agricultural and Food Centre (NPPC) for the Protection of Animals Used for Scientific and Educational Purposes (approval No. EK NPPC 05/2025). The Committee concluded that semen collection from bucks performed within this study constituted a non-experimental agricultural procedure and therefore did not require additional approval from the State Veterinary and Food Administration of the Slovak Republic.

Results

Volume, concentration and motility of fresh sperm

The average volume of fresh sperm obtained from the studied goats was 0.90 ± 0.23 ml. The average concentration of fresh sperm was $4.04 \pm 2.80 \times 10^9$ ml⁻¹. The overall

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motility in our study reached an average value of 88.94 % and the average progressive motility was 84.07 %.

Effect of diluent type on sperm motility after thawing

The cryopreservation process significantly affected the progressive sperm motility, as mostly the progressive motility of fresh sperm was higher than the measured progressive motility values, which were evaluated after thawing in both extenders. However, the effects of cryopreservation were not the same for both extenders. The percentages of progressive sperm motility (Table 1) after thawing were significantly ($p < 0.05$) higher when using OptiXcell® (65.38%) than when using Triladyl® (40.06%) extenders.

Table 1: The sperm motility after thawing using different extenders

Extender	Triladyl		OptiXcell	
Freezing type	MF (%)	PF (%)	MF (%)	PF (%)
PM	40.06 ± 6.66 ^b	36.85 ± 8.32 ^b	65.38 ± 8.90 ^a	57.13 ± 18.17 ^a

^{a,b} Values within rows with different superscripts are different; $p < 0.05$. Abbreviations: PM, progressive motility; MF, manual freezing; PF, programmable freezing.

Effect of freezing technique on sperm motility after thawing

The results show no statistically significant difference in the use of programmable and manual freezing when using Triladyl, indicating their mutual homogeneity. There was also no significant difference in the use of programmable and manual freezing techniques, when using the Optixcell sperm extender, indicating similar values within these two groups.

Discussion

The average fresh sperm volume values in our study were lower and concentrations - higher compared to the study on Malya and Boer goats (KASHOMA et al., 2025). The results of the study on male Korean Native Black Goats and Crossbred Goats (EUN-DO et al., 2025), where the collection was performed by electroejaculation, also show lower sperm volumes in males and higher concentrations compared to our results. The volumes and concentrations in our study were similar to those observed on Saanen goats (NISFIMAWARDAH et al., 2025). The sperm concentrations in our study were also similar to those concentrations in Gulabi goats (MUHAMMAD et al., 2025).

Total motility of fresh sperm in our study was similar to that of Gulabi goats (MUHAMMAD et al., 2025) and higher than that of Saanen goats (NISFIMAWARDAH et al., 2025). The progressive motility of fresh sperm in our

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study was higher than that of Boer, Saanen and Anglo-Nubian goats in the study of HANIFI et al. (2025) but lower compared to Malya and Boer goats in the study of KASHOMA et al. (2025).

The progressive motility after thawing in our results was higher compared to the study on Saanen goats using an Optixcell extender (NISFIMAWARDAH et al., 2025). The motility in our study was also significantly higher compared to the study on rams (SETIADI et al., 2025). Total motility after thawing was approximately the same as in Gulabi goats using the extender Triladyl (MUHAMMAD et al., 2025). VOZAF et al. (2021) achieved progressive motility values of 74.8% with Triladyl at 6-hour equilibration, which are higher values than in our study.

Although cryopreservation is the most important procedure for success in assisted reproductive biotechnology, this process causes significant structural and molecular changes in mammalian sperm (YANEZ-ORTIZ et al., 2021). Cryopreservation strongly alters acrosome integrity, destroys sperm function and alters membrane fluidity, phospholipid and protein patterns, all of which are associated with a decrease in enzymatic activity, motility, viability and the ability of sperm to fertilize an egg (SHADAN et al., 2004). The decrease in sperm viability after freezing is associated with intracellular changes after cryopreservation, leading to the formation of intracellular ice crystals that cause sperm membrane destruction, increase permeability and, ultimately, sperm death (KHAN et al., 2021).

OptiXcell® resulted in higher post-thaw progressive sperm motility than Triladyl®. No significant differences were observed between manual and programmable freezing. However, the small sample size ($n = 5$) and evaluation of motility alone limit the generalizability of the findings. Manual freezing may therefore be a useful alternative when programmable equipment is unavailable, but further studies with more animals and additional sperm quality parameters are needed. The main limitation of the present study is the relatively small number of bucks included ($n = 5$), which may limit the generalisability of the findings and the ability to fully account for inter-individual variability in semen characteristics and response to cryopreservation.

Nevertheless, the experimental design allowed repeated evaluation of semen samples under standardised laboratory conditions and provided preliminary information on the effects of the investigated cryopreservation manipulations. Future studies involving a larger and more diverse population of bucks are warranted to confirm the present findings and to better characterise individual variability in response to the cryopreservation protocol.

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DOI: <https://doi.org/10.59913/dagr.2026.21976>

References

- ARI, U.C. – YILDIZ, S. (2018): Storage of Goat Semen. Türkiye Klinikleri.
- ARIYAN, F. – FARZINPOUR, A. – FARSHAD, A. – SHARIFI, A. (2025): Protective Effects of *Lactobacillus plantarum* Secretions on Goat Sperm Quality During Cryopreservation. Biopreservation and Biobanking. <https://doi.org/10.1177/19475535251395868>
- BERČÍK, J. – VOZAF, J. – BALÁŽI, A. – CHRENEK, P. (2026): Cryopreservation of goat sperm: Effect of manual versus programmable freezing and different equilibration lengths on progressive motility. In Winter School – Animal Biotechnology 2026. SUA in Nitra. 78 p. <https://doi.org/10.15414/2026.9788055229416>
- EUN-DO, L. – MAYANGE, T.B. – DONG-KYO, K. – GA-EUN, K. – SANG-RAE, C. – KWAN-WOO, K. (2025): A Study on the Frozen Semen and Artificial Insemination Efficiency of Korean Native Black Goats and Crossbred Goats. Journal of the Korea Academia-Industrial cooperation Society, 26(1), 47–52. <https://doi.org/10.5762/KAIS.2025.26.1.47>
- HAFIDHA, F.G. – UTOMO, B. – SOEHARSONO – LESTARI, T.D. – HIDAJATI, N. – HERMADI, H.A. (2025): The Effect of Kemangi Leaf (*Ocimum Basilicum*) Extract in The Egg Yolk Skim Milk Diluent on The Plasma Membrane and Motility of Boerawa Goats' Sperm. Media Kedokteran Hewan, 36(3), 273–281. <https://doi.org/10.20473/mkh.v36i3.2025.273-281>
- HANIFI, H. – UMAR, S. – HANDARINI, R. – MAULANA, T. (2025): Kinematic Characteristics of Spermatozoa in Different Goat Breeds Using Computer Assisted Semen Analysis (CASA). Jurnal Agripet, 25(2), 209–213. <https://doi.org/10.17969/agripet.v25i2.47621>
- CHRENEK, P. – MAKAREVICH, A. – KUBOVIČOVÁ, E. – BULLA, J. – SUPUKA, P. (2025): Slovenské národné plemená zvierat. Slovenská poľnohospodárska univerzita. 99 p.
- KASHOMA, I.P. – KELLER, W.S. – KICHUKI, M.M. (2025): Comparative Efficacy of Soybean Milk and OptiXcell Extenders for Cryopreservation of Goat Semen. Tanzania Veterinary Journal, 40(1). <https://doi.org/10.4314/tvj.v40i1.6>
- KHAN, I.M. – CAO, Z. – LIU, H. – KHAN, A. – RAHMAN, S.U. – KHAN, M.Z. – SATHANAWONGS, A. – ZHANG, Y. (2021): Impact of Cryopreservation on Spermatozoa Freeze-Thawed Traits and Relevance OMICS to Assess Sperm Cryo-Tolerance in Farm Animals. Frontiers in Veterinary Science, 8. <https://doi.org/10.3389/fvets.2021.609180>
- KUMAR, A. – KANAKA, K.K. – MEHTA, H. – SINGH, G. – THAKUR, N.S. (2025): A Review of Current Challenges, Influential Factors, and Advancements in Goat Semen Cryopreservation. Archives of Current Research International, 25(3), 398–411. <https://doi.org/10.9734/acri/2025/v25i31131>

DOI: <https://doi.org/10.59913/dagr.2026.21976>

- LONE, S.A. – PRASAD, J.K. – GHOSH, S.K. – DAS, G.K. – BALAMURUGAN, B. – VERMA, M.R. (2018): Study on correlation of sperm quality parameters with antioxidant and oxidant status of buffalo bull semen during various stages of cryopreservation. *Andrologia*, 50(4). <https://doi.org/10.1111/and.12970>
- MOHAMMED, Z.A. – OMAR, C. (2025): Effect of Various Levels of Sage (*Salvia officinalis*) Essential Oil Drenching on Some Semen Parameters in Different Periods of Saanen Goat Bucks. *Basrah Journal of Agricultural Sciences*, 38(2), 455–466. <https://doi.org/10.37077/25200860.2025.38.2.34>
- MUHAMMAD, T. – KALWAR, Q. – RAHIMOON, M.M. – SOOMRO, H. – HUSSAIN, F. – LAGHARI, S.A. – SABOOR, A. – KOREJO, R.A. (2025): Effects of Triladyl® Extender and Vitamin E on Fresh and Frozen Thawed Semen of Gulabi Bucks. *Planta Animalia*, 4(5), 445–450. <https://doi.org/10.71454/PA.004.05.0250>
- NISFIMAWARDAH, L. – HUSEIN, F. – ASTUTI, E. – MONASDIR, M. – RISKAYANTI, R. – NUGROHO, A.A. (2025): Utilization of Various Diluents on the Quality of Frozen Semen from Saanen Goats. *Jurnal Ilmiah Peternakan Terpadu*, 13(1), 179–193. <https://doi.org/10.23960/jipt.v13i1.p179-193>
- RANJAN, R. – KUMAR, M. – GANGAWAR, C. – KHARCHE, S.D. (2021): Developments in Goat Semen Cryopreservation. *Animal Reproduction Update*, 1(1), 41–45. <https://doi.org/10.48165/aru.2021.1205>
- SETIADI, D.R. – MAGFIRA – RIWU, R.M.J. – ARIFIANITINI, R.I. (2025): Effectiveness of various commercial extenders in cryopreserving semen of local indonesian rams. *Jurnal Bioteknologi & Biosains Indonesia*, 12(1), 68–77. <https://doi.org/10.55981/jbbi.2025.9208>
- SHADAN, S. – JAMES, P.S. – HOWES, E.A. – JONES, R. (2004): Cholesterol Efflux Alters Lipid Raft Stability and Distribution During Capacitation of Boar Spermatozoa. *Biology of Reproduction*, 71(1), 253–265. <https://doi.org/10.1095/biolreprod.103.026435>
- SIAS, B. – FERRATO, F. – PELLICER-RUBIO, M.T. – FORGERIT, Y. – GUILLOUET, P. – LEBOEUF, B. – CARRIERE, F. (2005): Cloning and seasonal secretion of the pancreatic lipase-related protein 2 present in goat seminal plasma. *Biochimica et Biophysica Acta*, 1686(3), 169–180. <https://doi.org/10.1016/j.bbalip.2004.09.008>
- SIKKA, S.C. (2004): Role of oxidative stress and antioxidants in andrology and assisted reproductive technology. *Journal of Andrology*, 25(1), 5–18. <https://doi.org/10.1002/j.1939-4640.2004.tb02751.x>
- TRAN, K.T.T. – LU, S.N.C. – NGUYEN, D.L.K. (2025): Curcumin as a protective agent for goat sperm viability and DNA integrity against bisphenol A toxicity and cryopreservation stress. *Veterinary Research Communications*, 49(6). <https://doi.org/10.1007/s11259-025-10878-7>
- VOZAF, J. – MAKAREVICH, A. – BALÁŽI, A. – VASICEK, J. – SVORADOVÁ, A. – OLEXÍKOVÁ, L. – CHRENEK, P. (2021): Cryopreservation of ram semen:

DOI: <https://doi.org/10.59913/dagr.2026.21976>

Manual versus programmable freezing and different lengths of equilibration.

Animal Science Journal, 92(1). <https://doi.org/10.1111/asj.13670>

YANEZ-ORTIZ, I. – CATALAN, J. – RODRIGUEZ-GIL, J.E. – MIRO, J. – YESTE, M. (2021): Advances in sperm cryopreservation in farm animals: Cattle, horse, pig and sheep. Animal Reproduction Science, 246. <https://doi.org/10.1016/j.anireprosci.2021.106904>

Evaluation of some morphological measurements of Hungarian Hucul horse mares

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Abstract

This study evaluated four key body measurement traits in Hungarian Hucul mares: height at withers by stick and tape, heart girth, and cannon bone circumference. Data from 623 Hungarian Hucul mares collected between 2002 and 2025 were analysed using a linear animal model including sire line, mare family, year and age as fixed effects and animal as a random additive genetic effect. Genetic parameters (heritability, genetic and phenotypic correlations) were estimated utilizing the REML method via VCE-6 software. The descriptive statistical analysis was performed using the R statistical software. Heritability (h^2) values were above 0.7 for height at withers measured by stick and tape, as well as for heart girth size, while a lower value (0.45) was found for cannon bone circumference. The genetic correlation (r_G) between height at withers measured by stick and heart girth was 0.45, and, as expected, the highest (0.97) was observed between height at withers measured by stick and tape. The lowest phenotypic correlation (r_P) was 0.39, observed between height at withers measured by stick and cannon bone circumference, while the highest value (0.90) was also found between the two withers height measurements. Overall, while the Hungarian Hucul population shows relatively low phenotypic variability in the investigated body measurements, the moderate to high heritability of these traits suggests that corrective mating plans utilizing genetic information can be highly efficient for future breed management.

Keywords: Hucul horses, conformation, genetic parameters

Introduction

The Hucul horse breed originated in the north-eastern regions of the forested Carpathians. Following the Second World War, only a small number of animals survived, and the regeneration of the breed in Hungary was initiated by Csaba Anghy at the Budapest Zoo (MIHÓK, 2014). From those initial few mares and stallions, the current registered breeding population has grown to over 600 broodmares. Furthermore, all seven recognized sire lines are represented, comprising more than 60 breeding stallions in Hungary. Other substantial and genetically significant populations of the breed are located in Poland, Romania, and Slovakia (POSTA et al., 2020; Database of the National Association of Pony and Small Horse Breeders, 2025).

Since 1992, the National Association of Pony and Small Horse Breeders has been responsible for the maintenance of the breed (MIHÓK et al., 2019).

Before entering the studbook, animals must undergo a conformation evaluation, which includes the recording of specific body measurements. The conformation scoring utilizes a 100-point system, evaluating individual criteria on a scale of 0 to 10. A score below 4 in any criterion disqualifies the horse from breeding. For mares, the desirable optimal body measurements are as follows: 136-142 cm for height at withers measured by Lydtin stick, 146-152 cm for height at withers measured by tape, 160-176 cm for heart girth, and 17-18 cm for cannon bone circumference. Additionally, a minimum overall conformation score of 60 points is required (BREEDING PROGRAM OF HUCUL HORSES, 2020).

The objective of this study was to analyse the body measurements of the Hungarian Hucul mare population. This includes examining the age of entry into the studbook, identifying differences across individual years, sire lines, and mare families, as well as estimating the genetic parameters (heritability, genetic and phenotypic correlations) that can be calculated for these traits. The results of our analysis will establish an objective scientific basis and enable the breeding organization to determine further breeding directions. Quantifying these parameters can assist the breed's organization and provide useful information to breeders for preparing their mating plans.

Material and methods

Data for this study were obtained by digitizing the conformation scoring sheets of which 623 mares had complete records for all body measurements and were therefore included in the analysis. The dataset was kindly provided by the National Association of Pony and Small Horse Breeders. To support the genetic analysis, pedigree information was compiled using Microsoft Access, tracing the lineages

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back to the founder animals, resulting in a comprehensive pedigree file of 1,888 individuals.

The phenotypic evaluation focused on four key morphological traits:

Height at withers by stick (HWS), Height at withers by tape (HWT), Heart girth (HG), Cannon bone circumference (CBC)

To estimate the variance components and genetic parameters, the following linear model was applied:

$$Y_{ijklm} = \mu + Sireline_i + Marefamily_j + Year_k + Age_l + Animal_m + e_{ijklm}$$

Y_{ijklm}	=	measured trait;
μ	=	population mean;
$Sireline_i$	=	sire line of the mare;
$Marefamily_j$	=	mare family of the mare;
$Year_k$	=	effect of mare test's year;
Age_l	=	effect of age at measurement;
$Animal_m$	=	random effect of the mare;
e_{ijklm}	=	random residual term.

The descriptive statistical analysis was performed using the R statistical software (R CORE TEAM, 2025). The estimation of genetic parameters (heritability and correlations) was carried out utilizing the Restricted Maximum Likelihood (REML) method, implemented via the VCE-6 software package (GROENEVELD et al., 2008).

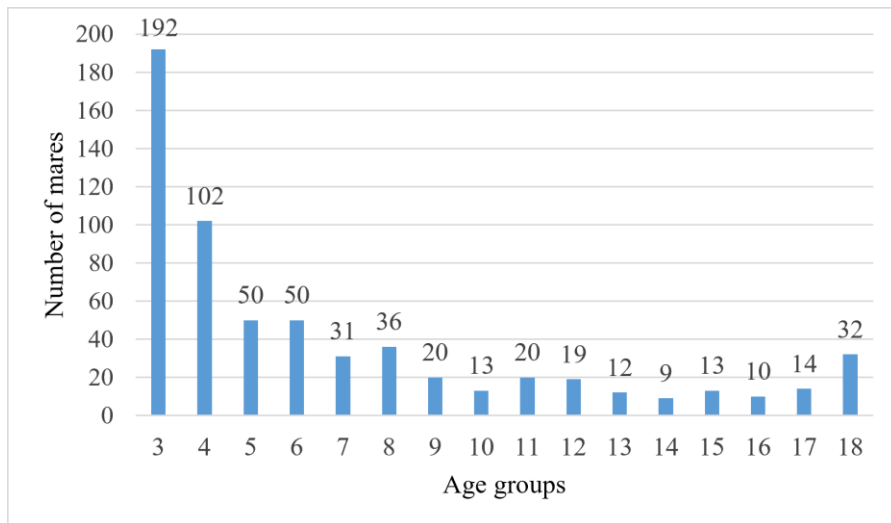


Figure 1: Number of Hucul mares by age (years)

Figure 1 shows the number of mares by age. The 3-year-old and 4-year-old age groups have the most individuals.

Results and discussion

The fixed effects included in the model were significant for each measurement trait. In the followings, the effect of sire lines and mare families will be shown.

Figure 2 illustrates the distribution of mare families within the evaluated population. Currently, Hungary possesses 27 Hucul mare families. The two largest families are of Hungarian origin: Aspiráns (14%) and Árvácska (20%). Several families are represented by a negligible percentage (0%), indicating that only 1-2 individuals belong to them.

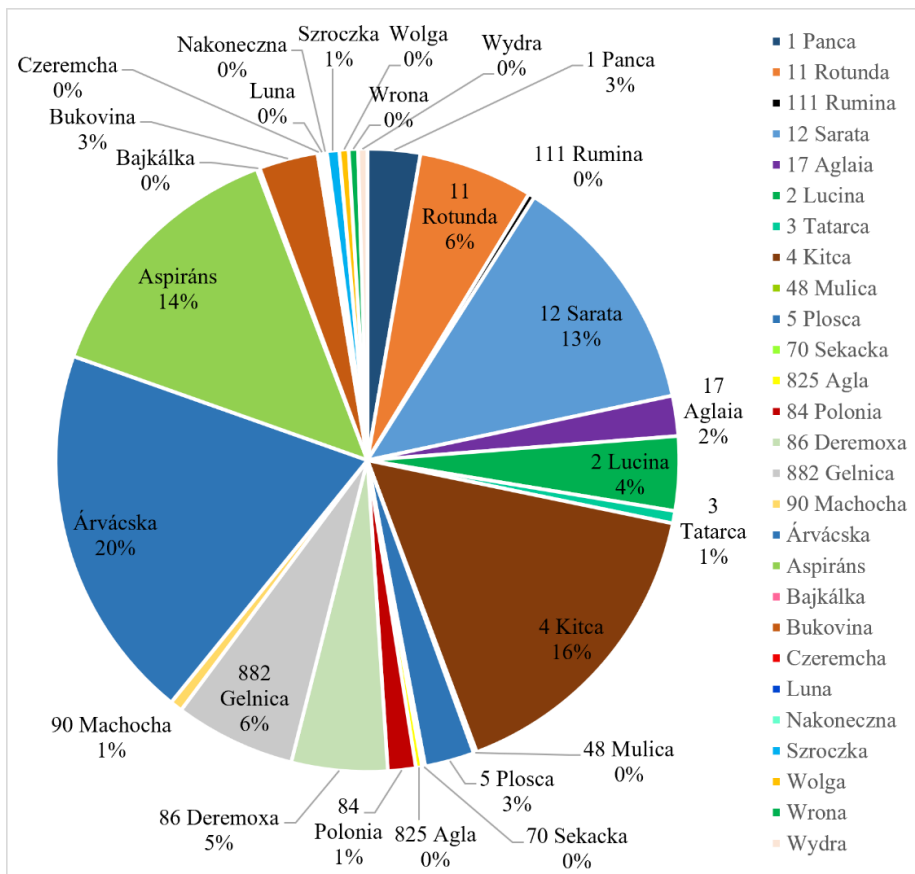


Figure 2: Distribution of Hucul mare families

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Furthermore, all seven recognized sire lines of the Hucul breed are present in Hungary (Figure 3.). The Hroby line represents the largest proportion of the population (24%), whereas the Gurgul line has the smallest representation (6%).

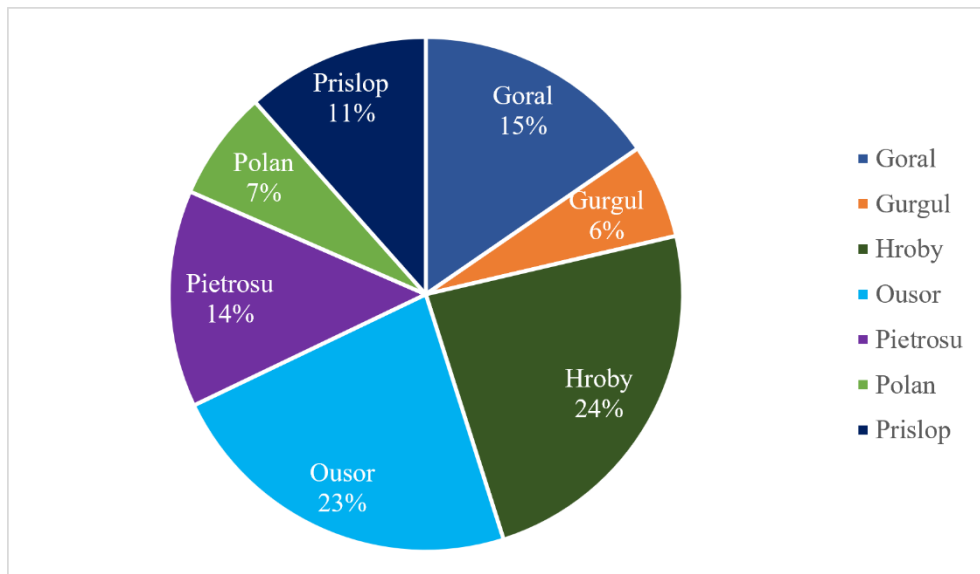


Figure 3: Distribution of Hucul sire lines

Table 1 summarizes the mean values, standard deviations, and coefficients of variation for each investigated trait of Hucul mares. Most of the measured values are between the desirable optimal measurements, although the average heart girth and cannon bone circumference slightly exceed the optimum.

Table 1: Descriptive statistics of body measurements in Hucul mares

Body measurements	Mean	SD	CV%
Height at withers (stick)	138.16 cm	3.88	2.81
Height at withers (tape)	146.41 cm	4.52	3.09
Heart girth	177.69 cm	10.23	5.76
Cannon bone circumference	18.02 cm	0.83	4.59

SD – standard deviation; CV% - coefficient of variation

TEIXEIRA et al. (2025) evaluated genetic parameters of morphological traits in Mangalarga Marchador horses. This native Brazilian riding horse breed has similar heart girth (172 cm) and cannon bone circumference (18 cm) values. KOMOSA and PURZYC (2009) reported similar body measurements for Hucul horses, with a mean

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height at withers measured by stick of 138.1 cm, heart girth of 177.5 cm, and cannon bone circumference of 19.1 cm, which are generally comparable to the values obtained in the present study.

The following tables contain descriptive statistics of each body measurement according to sire lines.

Table 2 presents the descriptive statistics of height at withers measured by stick across the different sire lines. The results indicate that individuals of the Prislop sire line have the highest average value (141.04 cm). In contrast, the Polan sire line represents the smallest individuals on average, with a mean height of 137.90 cm.

Table 2: Descriptive statistics of height at withers (by stick, in cm)

Sire line	N	Least-squares means	SE	lower CL	upper CL
Goral	96	139.02	0.64	137.77	140.28
Gurgul	37	138.63	0.81	137.04	140.22
Hroby	148	139.36	0.62	138.14	140.58
Ousor	142	138.87	0.64	137.62	140.11
Pietrosu	85	140.04	0.70	138.67	141.41
Polan	43	137.90	0.78	136.36	139.44
Prislop	72	141.04	0.71	139.65	142.43

SE – standard error; CL – 95% confidence level

Table 3 shows the descriptive statistics for height at withers measured by tape. Consistent with the stick measurements, the Prislop sire line had the largest average value (149.91 cm). The average HWT of the Pietrosu and Prislop sire lines is over 149 cm. The lowest average value was observed in the Polan sire line (146.54 cm).

Table 3: Descriptive statistics of height at withers (by tape, in cm)

Sire line	N	Least-squares means	SE	lower CL	upper CL
Goral	96	147.77	0.73	146.33	149.21
Gurgul	37	147.47	0.93	145.64	149.30
Hroby	148	148.18	0.71	146.78	149.58
Ousor	142	147.74	0.73	146.31	149.18
Pietrosu	85	149.02	0.80	147.45	150.60
Polan	43	146.54	0.90	144.77	148.31
Prislop	72	149.91	0.81	148.32	151.51

SE – standard error; CL – 95% confidence level

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Table 4. details the descriptive statistics of heart girth measurements according to sire lines. The Prislop line again demonstrates the largest average size (184.59 cm). Interestingly, the smallest average heart girth belongs to the Gurgul line (176.08 cm), closely followed by the Polan line (176.72 cm).

Table 4: Descriptive statistics of heart girth (in cm)

Sire line	N	Least-squares means	SE	lower CL	upper CL
Goral	96	179.30	1.67	176.02	182.59
Gurgul	37	176.08	2.13	171.91	180.26
Hroby	148	181.24	1.63	178.05	184.44
Ousor	142	180.85	1.67	177.58	184.12
Pietrosu	85	181.61	1.83	178.01	185.20
Polan	43	176.72	2.06	172.68	180.76
Prislop	72	184.59	1.85	180.95	188.23

SE – standard error; CL – 95% confidence level

Table 5 displays the descriptive statistics for cannon bone circumference across the sire lines. Aligning with the other body measurements, the Prislop sire line has the highest average thickness (18.26 cm). Below 18 cm there are individuals of the Polan and Ousor sire lines. The Polan sire line presents the lowest average value for this trait (17.93 cm).

Table 5: Descriptive statistics of cannon bone circumference (in cm)

Sire line	N	Least-squares means	SE	lower CL	upper CL
Goral	96	18.08	0.14	17.81	18.36
Gurgul	37	18.24	0.18	17.89	18.59
Hroby	148	18.15	0.14	17.88	18.42
Ousor	142	17.97	0.14	17.69	18.24
Pietrosu	85	18.18	0.15	17.87	18.48
Polan	43	17.93	0.17	17.59	18.27
Prislop	72	18.26	0.16	17.96	18.57

SE – standard error; CL – 95% confidence level

Table 6 presents the estimated heritabilities (on the diagonal), genetic correlations (above the diagonal), and phenotypic correlations (below the diagonal) for the evaluated body measurements. Heritability (h^2) estimates were high for height at withers measured by stick (0.73), height at withers by tape (0.70), and heart girth (0.73). However, a moderate heritability (0.45) was observed for cannon bone circumference. Regarding genetic correlations (r_G), the strongest relationship was

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found between the two withers height measurements (0.97), while the lowest was between height at withers by stick and heart girth (0.45). Similarly, the highest phenotypic correlation (r_p) was observed between the two height measurements (0.90), whereas the lowest (0.39) was recorded between height at withers by stick and cannon bone circumference.

Table 6: Estimated heritabilities and correlations for body measurements

	1	2	3	4
1 Height at withers (stick)	0.73	<i>0.97</i>	<i>0.45</i>	<i>0.50</i>
2 Height at withers (tape)	0.90	0.70	<i>0.62</i>	<i>0.67</i>
3 Heart girth	0.42	0.58	0.73	<i>0.72</i>
4 Cannon bone circumference	0.39	0.49	0.48	0.45

heritabilities on the diagonal (bold), genetic correlations above the diagonal (italic), and phenotypic correlations are below the diagonal

When comparing our findings to the literature, TEIXEIRA et al. (2025) reported lower heritability values (0.29–0.50) for morphological traits in Mangalarga Marchador horses. In contrast, VOSTRY et al. (2017) calculated a similar h^2 value for cannon bone circumference (0.427) in three Czech Draught Horse Breeds. JÖNSSON et al. (2014) calculated 0.55 h^2 value for CBC. For withers height, lower heritability estimates were reported by JÖNSSON et al. (2014) in Swedish Warmblood horses (0.67) and VICENTE et al. (2014) in Lusitano horses (0.61). ZIADI et al. (2025) estimated a comparable h^2 value for HWS in Pura Raza Española horses (0.76), though their estimates for HG and CBC values were different, 0.57 and 0.33. MOLINA et al. (1999) estimated 0.58 for height at withers, for HG a moderate value of 0.48, for CBC 0.35 in the Andalusian horse.

Regarding correlations, POSTA and KOMLÓSI (2007) measured r_p and r_G in Hungarian Sporthorses. Between HWS and HWT the r_p value was 0.97. All other phenotypic correlations were stronger. Between HWS and HG, CBC and between HWT and HG the r_G values were similar. Other r_G values in sporthorses were weaker. VOSTRY et al. (2017) also found similar genetic correlation values, except for the relationship between cannon bone circumference and heart girth (0.359).

These differences in the literature can be explained by the fact that the traits were evaluated across different breeds, populations, and environmental conditions. Nevertheless, the moderate to high heritability coefficients observed in our study suggest that corrective mating for conformation and type characteristics, utilizing genetic information, can be highly efficient in the Hungarian Hucul population.

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Conclusion and recommendation

The investigated body measurements showed relatively low phenotypic variability in the Hungarian Hucul mare population. The highest mean values were generally observed in the Prislop sire line, whereas the lowest mean values were generally observed in the Polan sire line. These findings may have practical applications in breed management. If a breeder aims to adjust the size of their horses (whether to increase or decrease the overall frame in relation to the breed's optimal expectations) the descriptive phenotypic data provided in this study can serve as a valuable tool for designing targeted mating plans.

The moderate to high heritability estimates indicate that the investigated traits may respond to selection and could therefore be considered in corrective mating strategies. The very high genetic correlation between stick and tape measured height at withers indicates that the two measurements are largely controlled by the same genetic factors. Additionally, heart girth and cannon bone circumference showed a relatively high genetic correlation. Overall, integrating these genetic parameters with the current sire line statistics will assist both the breeding organization and individual breeders in maintaining and improving the optimal conformation of the Hucul breed.

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References

- BREEDING PROGRAM OF HUCUL HORSES (A hucul lófajta tenyésztési programja) (2020): https://www.ponieskislo.hu/_data/_vfs/hucul-tenyprog-2020.pdf
- GROENEVELD, E. – KOVAČ, M. – MIELENZ, N. (2008): VCE User's Guide and Reference Manual, Version 6.0. Institute of Farm Animal Genetics, Friedrich-Loeffler-Institut (FLI), Neustadt, Germany.
- JÖNSSON, L. – NÄSHOLM, A. – ROEPSTORFF, L. – EGENVALL, A. – DALIN, G. – PHILIPSSON, J. (2014): Conformation traits and their genetic and phenotypic associations with health status in young Swedish warmblood riding horses. *Livestock Science*, 163: 12 – 25. <https://doi.org/10.1016/j.livsci.2014.02.010>

DOI: <https://doi.org/10.59913/dagr.2026.23885>

- KOMOSA, M. – PURZYC, H. (2009): Konik and Hucul horses: A comparative study of exterior measurements. *Journal of Animal Science*, 87(7): 2245–2254. <https://doi.org/10.2527/jas.2008-1501>
- MIHÓK S. (2014): A hucul. Magyar Lótenyésztők Országos Szövetsége és Mezőgazda Kiadó. Budapest. 252 p.
- MIHÓK S. – POSTA J. – SOMOGYVÁRI E. (2019): A Kárpátok kislova, a hucul ló populációgenetikai jellemzése. Póni és Kislótenyésztők Országos Egyesülete, Debrecen. 112p.
- MOLINA, A. – VALERA, M. – DOS SANTOS, R. – RODERO, A. (1999): Genetic parameters of morphofunctional traits in Andalusian horse. *Livestock Production Science*, 60(2-3): 295–303. [https://doi.org/10.1016/S0301-6226\(99\)00101-3](https://doi.org/10.1016/S0301-6226(99)00101-3)
- POSTA, J. – KOMLÓSI, I. (2007): Genetic analysis of selected body measurements of Hungarian Sport Horse mares. *Acta Agraria Debreceniensis*, 26: 40–43. <https://doi.org/10.34101/actaagrar/26/3052>
- POSTA, J. – SOMOGYVÁRI, E. – MIHÓK, S. (2020): Historical changes and description of the current Hungarian Hucul horse population. *Animals*, 10(7): 1242. <https://doi.org/10.3390/ani10071242>
- R CORE TEAM (2025): R: A Language and Environment for Statistical Computing. R Foundation for Statistical Computing, Vienna, Austria. <https://www.R-project.org>
- TEIXEIRA, C.S. – SANCLER-SILVA, Y.F.R. – RAMOS, P.V.B. – CARRARA, E.R. – DE PAIVA, J.T. – MARQUES, D.B.D. – NASCIMENTO, M. – NASCIMENTO, A.C.C. – E ALVES, T.T.B. – LOPES, P.S. – E SILVA, F.F. (2025): Genetic parameters for latent variables derived from factor analysis of morphological and gait traits in Mangalarga Marchador horses. *Livestock Science*, 295: 105691. <https://doi.org/10.1016/j.livsci.2025.105691>
- VICENTE, A.A. – CAROLINO, N. – RALÃO-DUARTE, J. – GAMA, L.T. (2014): Selection for morphology, gaits and functional traits in Lusitano horses: I. Genetic parameter estimates. *Livestock Science*, 164: 1–12. <https://doi.org/10.1016/j.livsci.2014.01.020>
- VOSTRY, L. – VOSTRÀ-VYDROVÁ, H. – HOFMANOVÁ, B. – VESELÁ, Z. – SCHMIDOVÁ, J. – MAJZLIK, I. (2017): Genetic parameters for linear type traits in three Czech draught horse breeds. *Agriculturae Conspectus Scientificus*, 82(2): 111–115.
- ZIADI, C. – DEMYDA-PEYRÁS, S. – VALERA, M. – PERDOMO-GONZÁLEZ, D. – LASECA, N. – RODRÍGUEZ-SAINZ DE LOS TERREROS, A. – ENCINA, A. – AZOR, P. – MOLINA, A. (2025): Comparative analysis of genomic and pedigree-based approaches for genetic evaluation of morphological traits in pura raza Española horses. *Genes*, 16(2): 131. <https://doi.org/10.3390/genes16020131>

Donkey (*Equus asinus*) welfare in contemporary husbandry systems

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Abstract

The welfare of donkeys (*Equus asinus*) in contemporary husbandry systems is shaped by the interaction of biological, behavioural, and environmental factors. Their evolutionary adaptation to arid environments has led to high metabolic efficiency and specific behavioural traits that may become risk factors under modern conditions. Abundant feeding and reduced activity often result in metabolic disorders such as obesity, insulin resistance, and hyperlipemia, increasing the risk of laminitis and compromised welfare. Assessment is complicated by their stoic nature, as donkeys rarely show clear signs of pain. Therefore, traditional evaluation methods are often insufficient, requiring objective approaches based on subtle behavioural changes, body condition, and physiological indicators. Integrated assessment enables early detection and prevention of chronic disorders. Environmental conditions are also critical. Despite their perceived resilience, donkeys are sensitive to moisture and low temperatures, while poor housing increases the risk of respiratory and hoof diseases. The shift from working roles to milk production, tourism, and therapy introduces additional challenges, including reduced activity and increased stress. Improving welfare requires species-specific management and better education of owners. In conclusion, donkey welfare depends on an integrated approach combining scientific knowledge, objective assessment, and management aligned with species-specific needs.

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Keywords: donkey welfare; *Equus asinus*; metabolic disorders; husbandry systems; welfare assessment

Introduction

The donkey (*Equus asinus*) has had a key influence on the development of human civilization as a primary labour force in transport and tillage. Despite their importance for economic stability, donkeys have been systematically neglected in scientific and social circles throughout history in favour of horses (MCLEAN et al., 2018). The misperception of donkeys as “small horses with big ears” leads to inappropriate treatment in feeding and care. Such an approach directly threatens their welfare under modern husbandry and breeding settings (THE DONKEY SANCTUARY, 2015). Animal welfare is today defined as a state in which an animal is healthy, nourished, safe and able to express its innate behaviour, without suffering pain or fear. This concept is most often applied through the “Five Freedoms” model, which integrates the physical health and emotional state of the individual (FAWAC, n.d.). The importance of protecting working animals is globally recognized, as highlighted in the FAO report on the role, impact and welfare of traction and transport animals (FAO, 2014). In developing countries, welfare is primarily impaired by overwork and inappropriate equipment. On the contrary, in developed parts of Europe, donkeys face metabolic diseases and obesity due to overfeeding (TUČKORIĆ, 2021). In the Republic of Croatia, indigenous donkey breeds represent an invaluable genetic heritage and have become key factors in environmental conservation and tourism. It is precisely this change in role that requires the adjustment of housing standards, since donkeys have a specific resistance to climatic conditions (OSTHAUS et al., 2018).

The aim of this research is to analyse the welfare factors of donkeys through the identification of their biological and ethological needs. Special emphasis is placed on the application of objective scientific parameters such as the Animal Welfare Indicators (AWIN) system and the definition of guidelines for improving care in order to prevent suffering of this species.

Welfare assessment in donkeys

Objective assessment of donkey welfare represents one of the most complex challenges in modern veterinary medicine and zootechnics. Donkeys have a unique behavioural repertoire that often masks states of severe pain or chronic stress, which requires the use of precise, scientifically based instruments (DALLA COSTA et al., 2021). The development of standardized systems has enabled the transition from momentary observation to quantitative analysis of parameters that encompass physical health, emotional state and the quality of the environment in which the

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animal resides. This approach eliminates guesswork and subjective assessments that have often been the cause of erroneous conclusions. The system is based on objective evidence collected during field research by the authors who have addressed this topic. According to research by MASEBO et al. (2025), the use of such indicators allows for the early recognition of metabolic and psychological deviations before they become irreversible. The application of the AWIN system represents a state-of-the-art approach to objective assessment, as it integrates direct observations of the animal with an assessment of housing conditions, focusing on proper nutrition, housing, health and appropriate behaviour (DALLA COSTA et al., 2021). The protocol is designed to be easily applied in different breeding settings. It provides clear guidelines for assessing welfare regardless of the breed or purpose of the donkeys. This specific model allows owners and veterinarians to recognize subtle signs of impaired welfare through the assessment of skin cleanliness, hooves and gait (DAI et al., 2016). The Donkey Grimace Scale (DGS) further facilitates the recognition of suffering (ORTH et al., 2020), and the application of this protocol has proven to be crucial because signs of pain in donkeys are often more difficult to detect than in horses. Each assessment requires a high level of observer education. FARIAS et al. (2021) emphasize that such systematic application is not only a diagnostic tool, but also a preventive mechanism that allows owners to timely adjust living conditions, thereby raising welfare standards in the long term. The Body Condition Scoring (BCS) system is a key method for indirect assessment of nutritional status, using a scale from 1 to 5 (THE DONKEY SANCTUARY, 2015). The assessment procedure should be carried out regularly at intervals of several weeks. In this way, trends in the animal's weight change can be more easily monitored. The implementation requires physical palpation of the neck, withers and loins, since MANDIĆ et al. (2025) point out that it is precisely palpation of the neck that reveals specific fat deposits that are a permanent indicator of obesity and metabolic stress. Since the ribs in donkeys are not often visible due to the thickness of the skin, palpation remains a necessary step to avoid erroneous assessments (TUČKORIĆ, 2021). Systematic condition monitoring prevents the development of subclinical inflammatory processes associated with obesity (MASEBO et al., 2025), while behavioural indicators, such as ear position and muscle tension around the eyes, serve as precise indicators of acute pain (DALLA COSTA et al., 2021). Visual monitoring of behaviour within the herd provides additional information about the psychological state. When animals show apathy or withdrawal from the herd, this is a clear sign of impaired welfare (ARTEMIOU et al., 2021). Elevated levels of cortisol in the hair confirm chronic stress in animals exposed to adverse husbandry conditions (BUKHARI et al., 2025), and by monitoring metabolic parameters such as urea and ACTH hormones (ZHAO et al., 2020), experts can make an accurate diagnosis and provide a comprehensive assessment of the condition of each individual. LÉNÁRT et al. (2017) draw attention to the frequent constitutional faults, organismal weaknesses in donkeys. A significant part of the population consists of

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individuals with the defects toeing out forelimb, sickle-hocked hind limb, and X-leggedness. Extreme physical deformities are also related to the well-being of the animals and affect their performance and lifespan.

Sustainable donkey husbandry

Understanding the welfare of donkeys in modern husbandry systems requires an analytical review of the interaction between their inherited biological mechanisms and the conditions imposed on them by humans. The greatest challenges in practice often arise from anthropocentric errors, where these animals are imposed with standards of housing developed for horses, which is unsustainable in the long term. As MASEBO et al. (2025) point out, ensuring adequate welfare is not only the absence of disease, but a comprehensive approach that includes nutritional, environmental and husbandry conditions adapted to the specific needs of this species. The differences in anatomy and physiology between these species are often ignored in everyday practice. According to research conducted by THE DONKEY SANCTUARY (2015), inadequate nutrition and exposure to adverse climatic conditions are the most common causes of chronic health impairment in European farms. An additional difficulty in assessment is represented by husbandry conditions, because environmental stressors directly affect the immune system and homeostasis of the organism (ZHAO et al., 2020). Feeding represents one of the greatest challenges in modern donkey husbandry due to their exceptional metabolic efficiency. Adapting to the specifics of conditions of constant scarcity, donkeys have developed a digestive system that can extract nutrients from very poor and fibrous forage, which makes them susceptible to serious disorders in conditions of abundance. The incidence of colic in older donkeys is often associated with dental diseases and the absence of molars. As stated by THIEMANN and SULLIVAN (2019), the incidence of colic regularly results in secondary hyperlipemia, which critically threatens the chances of a successful treatment outcome. According to research conducted by MANDIĆ et al. (2025), obesity in donkeys, and especially in dairy donkeys, is directly associated with subclinical metabolic disorders. Their research highlights that elevated levels of urea and uric acid in obese individuals can be used as early biomarkers of inflammatory processes that precede clinical symptoms of laminitis. To maintain optimal welfare, feeding must be based on voluminous forage with a high crude fibre content and strictly limited access to new grass (THE DONKEY SANCTUARY, 2015). When exposed to rain without adequate shelter, the donkey loses its ability to retain body heat, which leads to hypothermia and chronic physiological stress. The problem of inadequate drainage systems, highlighted in dairy husbandry by KOVANDŽIĆ et al. (2024), has serious consequences for the hooves. The resting area must be clean and spacious enough to allow all individuals to lie on dry bedding. There is a clear correlation between the weight of the load carried by the donkey and the concentration of cortisol in its hair, which confirms chronic stress (BUKHARI et al., 2025). ZHAO ET AL (2020) found

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that long-term transport drastically changes the composition of the nasal microbiota and increases the level of the hormone ACTH, while JIANG et al. (2021) prove that transport stress negatively affects the intestinal microflora. The introduction of standardized assessment protocols is therefore a key factor in the long-term improvement of living conditions (FSAHAYE et al., 2018). The status of donkeys in Croatia is deeply connected to cultural identity, but mechanization has led to the threat of indigenous breeds. The change in the purpose of breeding has brought new risks, with the transition to dairy systems requiring new standards adapted to the reduction of the animal's physical activity (MASEBO et al., 2025). The change in the animal's primary function often results in reduced supervision by the owner. In indigenous breeds, the risk of metabolic disorders that impair their welfare increases (MANDIĆ et al., 2025), while adequate drainage is often lacking on dairy farms (KOVANDŽIĆ et al., 2024). Stress caused by poor housing or rough treatment inhibits the milk release reflex, and tourist use without rest additionally exhausts the animals (TUČKORIĆ, 2021). The legislative framework provides basic protection, but hidden signs of impaired welfare often remain beyond the reach of inspection. Lack of understanding of specific ethology is a major cause of subclinical diseases, therefore standardized assessment programs play a key role in raising standards (ARTEMIOU et al., 2021). In addition to regulations that condition incentives on compliance with zootechnical standards (ANIMAL PROTECTION LAW, 2017, 2019), systems such as AWIN need to be introduced to enable more objective assessment (DALLA COSTA et al., 2021).

Conclusion and recommendation

Research to date clearly indicates that moving away from the traditional, anthropocentric identification of donkeys with horses is a key prerequisite for achieving an appropriate level of their welfare. The specific physiology, extremely energy-efficient metabolism and characteristic stoic way of expressing pain require completely different zootechnical standards from those applied in horse breeding. It is precisely this stoicism that represents the greatest diagnostic challenge, because the ability to conceal pain often leads to late recognition of serious health conditions, which makes it necessary for the veterinary profession and breeders to apply objective assessment methods. In modern breeding, and especially in the Republic of Croatia, we are witnessing a significant change in the role of the donkey from an exclusively working animal to an animal for dairy and tourist purposes, which brings new risks. Obesity, insulin resistance and laminitis are becoming dominant threats to welfare due to inadequate energy-rich nutrition and reduced physical activity, which is why the systematic application of scientifically based systems, such as the AWIN system and BCS scoring, is necessary, which enable early detection of impaired welfare through subtle changes in facial expression and analysis of body

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condition. Improving the welfare of donkeys in Croatia must not rely solely on strict legal frameworks, but primarily on continuous and professional education of owners, which is key to recognizing the specific needs of the species. A deep understanding of the fact that donkeys require dry shelter due to their specific hair structure, fibre-based nutrition and the necessary preservation of social bonds through the phenomenon of permanent pair bonding, is the only way to sustainable breeding. Only by working together with scientific knowledge, objective indicators and an educated ethical approach can we ensure the long-term survival and quality of life of these unique indigenous breeds in the future.

References

- ARTEMIOU, E. – HUTCHISON, P. – MACHADO, M. – ELLIS, D. – BRADTKE, J. – PEREIRA, M.M., CARTER, J. – BERGFELT, D. (2021): Impact of human-animal interactions on psychological and physiological factors associated with veterinary school students and donkeys. *Frontiers in Veterinary Science*, 8, Article 701302. <https://doi.org/10.3389/fvets.2021.701302>
- Animal Welfare Indicators. (2015): AWIN welfare assessment protocol for donkeys. https://doi.org/10.13130/AWIN_DONKEYS_2015
- BUKHARI, S.S.U.H. – LI, C.M. – KENÉZ, Á. – STEAGALL, P.V. – MCELLIGOTT, A.G. – PARKES, R.S.V. (2025): Donkey hair cortisol concentrations are associated with carrying heavy load and being beaten at work. *BMC Veterinary Research*, 21, Article 582. <https://doi.org/10.1186/s12917-025-05021-2>
- DAI, F. – DALLA COSTA, E. – MURRAY, L.M.A. – CANALI, E. – MINERO, M. (2016): Welfare conditions of donkeys in Europe: Initial outcomes from on-farm assessment. *Animals*, 6(1), Article 5. <https://doi.org/10.3390/ani6010005>
- DALLA COSTA, E. – DAI, F. – MURRAY, L.M.A. – CANNAS, S. – CANALI, E. – ZANELLA, A.J. – MINERO, M. (2021): The development of the AWIN welfare assessment protocol for donkeys. *Brazilian Journal of Veterinary Research and Animal Science*, 58 (Special Issue), Article e173333. <https://doi.org/10.11606/issn.1678-4456.bjvras.2021.173333>
- FARIAS, S. de S. – GAMEIRO, M.B.P. – MONTECHESE, A.C.D. – BERNARDINO, T. – OLIVEIRA, C.A. de A. – ZANELLA, A.J. (2021): Case report: The use of the AWIN welfare assessment protocol to monitor a group of abandoned donkeys. *Brazilian Journal of Veterinary Research and Animal Science*, 58, Article e174701. <https://doi.org/10.11606/issn.1678-4456.bjvras.2021.174701>
- FARM ANIMAL WELFARE ADVISORY COUNCIL [FAWAC] (n.d.): Animal welfare guidelines for horses, ponies and donkeys. Department of Agriculture, Food and the Marine.

DOI: <https://doi.org/10.59913/dagr.2026.24254>

FOOD AND AGRICULTURE ORGANIZATION OF THE UNITED NATIONS [FAO] (2014): The role, impact and welfare of working (traction and transport) animals: Report of the FAO–The Brooke Expert Meeting (Animal Production and Health Report No. 5).

FSAHAYE, S. – KUMAR, N. – KEBEDE, E. – ABEBE, N. (2018): Health and welfare assessment of working donkeys in and around Rama town, Tigray, Ethiopia. *Ethiop. Vet. J.*, 22(1):26–39. <https://doi.org/10.4314/evj.v22i1.3>

JIANG, G. – ZHANG, X. – GAO, W. – JI, C. – WANG, Y. – FENG, P. – FENG, Y. – ZHANG, Z. – LI, L. – ZHAO, F. (2021): Transport stress affects the fecal microbiota in healthy donkeys. *Journal of Veterinary Internal Medicine*, 35(5):2449–2457. <https://doi.org/10.1111/jvim.16235>

KOVANDŽIĆ, M. – PINTARIČ, Š. – ĐORĐEVIĆ, J. – LEDINA, T. – SAVIĆ RADOVANOVIĆ, R. – ČOBANOVIĆ, N. (2024): On-farm welfare conditions of dairy donkeys: A case study in Northern Serbia. *Meat Technology*, 65(2):146–153. <https://doi.org/10.18485/meattech.2024.65.2.8>

LÉNÁRT, Z. – ERNST, M. – GÁSPÁRDY, A. (2017): Preliminary results on body conformation of Hungarian Fallow Donkey. *Danubian Animal Genetic Resources*. 2:51–58. https://www.dagene.eu/docs/pazin_2017/dagene_pazin_2017.pdf

MANDIĆ, A. – MATORKIĆ, B. – SPARIOSU, K. – RADAKOVIĆ, M. – MITROVIĆ, A. – NENADOVIĆ, K. – KOVAČEVIĆ FILIPOVIĆ, M. (2025): Obesity, urea and uric acid: Potential indicators of subclinical metabolic imbalance in donkeys (Pilot study). *Pakistan Veterinary Journal*, 45(3):1415–1418. <https://doi.org/10.29261/pakvetj/2025.246>

MASEBO, N.T. – BENEDETTI, B. – ANGELONI, M.G. – LEE, L. – BIGI, D. – PADALINO, B. (2025): Systematic literature review on donkeys (*Equus asinus*): Husbandry and welfare in Europe. *Animals*, 15(19), Article 2768. <https://doi.org/10.3390/ani15192768>

MCLEAN, A.K. – NAVAS GONZÁLEZ, F.J. (2018): Can scientists influence donkey welfare? Historical perspective and contemporary view. *Journal of Equine Veterinary Science*, 65, 25–32. <https://doi.org/10.1016/j.jevs.2018.03.008>

Zakon o zaštiti životinja (Animal protection act) (2017, 2019): Narodne novine, Zagreb, Croatia.

ORTH, E.K. – NAVAS GONZÁLEZ, F.J. – IGLESIAS PASTRANA, C. – BERGER, J.M. – JEUNE, S.S.L. – DAVIS, E.W. – MCLEAN, A.K. (2020): Development of a Donkey Grimace Scale to Recognize Pain in Donkeys (*Equus asinus*) Post Castration. *Animals (Basel)*. 10(8):1411. <https://doi.org/10.3390/ani10081411>

OSTHAUS, B. – PROOPS, L. – LONG, S. – BELL, N. – HAYDAY, K. – BURDEN, F. (2018, June 28). Weathering the weather: Effects of the environment on donkey, mule and horse welfare [Conference presentation]. UFAW Animal

DOI: <https://doi.org/10.59913/dagr.2026.24254>

Welfare Conference: Recent Advances in Animal Welfare Science VI, Newcastle upon Tyne, United Kingdom.

THE DONKEY SANCTUARY. (2015): The donkey care handbook (4th ed.). The Donkey Sanctuary.

THIEMANN, A.K. – SULLIVAN, R.J.E. (2019): Gastrointestinal disorders of donkeys and mules. *Veterinary Clinics of North America: Equine Practice*, 35(3):419–432. <https://doi.org/10.1016/j.cveq.2019.08.001>

TUČKORIĆ, A. (2021): Metabolički poremećaji u magaraca i važnost magarećeg mlijeka u ljudskoj prehrani [Diplomski rad, Sveučilište u Zagrebu, Veterinarski fakultet]. Repozitorij Veterinarskog fakulteta Sveučilišta u Zagrebu.

ZHAO, F. – JIANG, G. – JI, C. – ZHANG, Z. – GAO, W. – FENG, P. – LI, H. – LI, M. – LIU, H. – LIU, G. – MAGALHAES, H.B. – LI, J. (2020): Effects of long-distance transportation on blood constituents and composition of the nasal microbiota in healthy donkeys. *BMC Veterinary Research*, 16, Article 338. <https://doi.org/10.1186/s12917-020-02563-5>

Growth potential of autochthonous wether lambs on natural pasture between May and October

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Abstract

The year 2026 has been declared the International Year of Rangelands and Pastoralists by the UNESCO. To understand the exact capabilities, this study examines the growing and fattening ability of sheep under extensive conditions during the primary plant-growing season (between mid-May and mid-October). The investigation included wether lambs of Hungarian native sheep breeds with German Mutton Merino (GM) control individuals (5 per breed). To ensure reliable comparison, the values of live weight and daily gain were adjusted for the assigned days of life then a one-way ANOVA was performed on them with the fixed effect of breed. Data preparation and processing were done with the software Statistica (version 14, TIBCO). The effect of the age of the individuals was not significant, which means that lambs of similar age participated in the fattening period. However, differences between breeds were already evident in the initial weight (as a result of the growth during pre-weaning period and the weight at maturity): the Tsigai, Merino, Racka and GM started the study with a significantly higher weight than the Black Racka and the Dairy Tsigai. The lambs gained weight in the first and last fattening months, but lost weight overall during the intervening three months. The extensive, pasture-based fattening showed that the acclimatization and mid-summer weight losses were regained in accordance with the compensatory capacity of the young animals. The initial difference between breeds remained in the final weight, and this is also reflected in the average daily gain during fattening.

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Keywords: native sheep breeds, whether fattening, pasture-based feeding system, compensatory growth.

Introduction

Local sheep breeds are increasingly valued not only as components of agricultural heritage, but also as reservoirs of genetic diversity that may support resilience, adaptation and product differentiation in changing production systems.

In Hungary, several sheep breeds that were historically important from the Middle Ages to the nineteenth century have lost their former economic dominance and are now maintained mainly as gene reserves and elements of cultural heritage (HANKÓ, 1937; BÖKÖNYI, 1974). Among the traditional Hungarian sheep populations, the Hungarian Sheep (Racka) and the Tsigai occupy a particularly important historical position. The Hungarian Sheep belongs to the Zackel-type sheep of Eastern and Central Europe and is morphologically distinguished by the long, twisted V-shaped horns present in both sexes (MATOLCSI, 1975; GAÁL, 1986; DUNKA and JÁVOR, 1991; DUNKA, 2006). The Tsigai is another long-established breed of major regional importance. According to the available breed descriptions, it originated in Asia Minor and reached the Carpathian Basin from the thirteenth century onward (SCHANDL, 1955; FÉSÜS, 1991). The Milking Tsigai represents a dairy-oriented Tsigai type maintained under Hungarian conditions. It is also of Balkan origin and established in Hungary by the end of the eighteenth century (GÁSPÁRDY et al., 2016). In contrast to these traditional populations, the Hungarian Merino represents the improved breed type that progressively displaced several indigenous and long-established sheep populations in Hungary. The modern Hungarian Merino was formed during the twentieth century from fine-wool Merino populations and local breeds, followed later by meat-oriented improvement (SZABÓ et al., 2016). The year 2026 has been declared the International Year of Rangelands and Pastoralists by the UNESCO.

To understand the exact capabilities, this study examines the growing and fattening ability of Hungarian autochthonous sheep breeds under extensive conditions during the primary plant-growing season (between mid-May and mid-October).

Material and methods

The investigation included wether lambs (5 per breed) of Hungarian native sheep breeds (Tsigai, Dairy Tsigai (DT), Hungarian Merino, as well as White and Black Rackas) with German Mutton Merino (GM) control individuals. The lambs had identifiers with the same unique individual life number in accordance with the EU

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regulations (Council Regulation (EC) No 21/2004) and additionally tattoos in the auricles. They were born in seed stocks with known origin. When selecting lambs, it was important that they were born from at least three rams of each breed. Bloodless castration with pliers Burdizzo style (VF301N, Aesculap, Inc., 3773 Corporate Parkway Center Valley, 18034 PA, USA) was performed in the first month after birth. For a competent farmer, to use this procedure without professional anaesthesia under 3 months of age (up to 90 days) is permitted in Europe. Therefore, no statement on animal ethics or welfare approval was requested for the castration.

The extensive fattening of the weaned wethers on pasture was carried out in the area of the Körös-Maros National Park (KMNP, in Dévaványa). After a short (9-day-long) acclimatization in their own barn on *hey ad libitum*, the lambs lived in natural environment, in an old-growth grassland, grazing freely during the day and spending the night in a sheep barn. One of the DT died after delivery with a diagnosis of pneumonia. They were not given any feed supplements. Lick salt and drinking water were always available to them. The pasture grass yield in the year of the study was about 1.8-1.9 tonnes per hectare (t/ha). This was considered exceptionally favourable in the area dominated by 80% of the alkaline meadow plant community. Here the most characteristic plant was the meadow foxtail (*Alopecurus pratensis*), but creeping bentgrass (*Agrostis stolonifera*), wand loosestrife (*Lythrum virgatum*) and birdsfoot deervetch (*Lotus corniculatus*) were also found in smaller quantities.

In addition to the delivery (May 10), initial (May 19) and final (October 14) weights of the lambs, their monthly live weights were also determined using a portable digital scale (SN: 206XM018, Iconix FX1, Iconix New Zealand Ltd., Oamaru, New Zealand) with an accuracy of 0.1 kg. The average daily gain was calculated by knowing the age and the weight gained per period between two consecutive weighting and during the entire fattening period.

To ensure reliable comparison, the values of live weight and daily gain were first adjusted for the assigned days of life. Then, to reveal breed differences a one-way ANOVA completed with Tukey HSD (Honestly Significant Difference) post-hoc test was used where the breed was considered as a fixed effect. Data preparation and processing were done with the software Statistica (version 14, TIBCO, 4980 Great America Parkway Santa Clara, CA 95054, USA).

Results and discussion

The effect of the age of the individuals in almost all the cases (per weight measurement) was not significant, which means that lambs of similar age participated in the fattening period. However, differences between breeds were already evident in the delivery weight as a result of the growth during pre-weaning period. The Tsigai, Merino, Racka and GM started the study with a significantly higher weight than the Black Racka and the Dairy Tsigai. During the acclimatization

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period, weight decreased, so daily gain took on negative values (Table 1). The average daily loss was 103 g. The only exception was Dairy Tsigai, which was able to maintain its gain during the acclimatization period.

Table 1: Delivery weight adjusted for 97 days of age (age $p=0.192$) and daily gain during acclimatization (age $p=0.584$) according to the studied sheep breeds

Breeds	n	Delivery weight, kg $p<0.001$	Std. Err.	Daily gain during acclimatization (between day 97 and 106), g $p=0.001$	Std. Err.
Tsigai	5	31.6 ^a	2.73	-237 ^b	155.6
Dairy Tsigai	5	21.4 ^{bc}	2.83	78 ^a	50.3
Hung. Merino	5	24.0 ^{bc}	2.03	-101 ^{ab}	97.3
White Racka	5	24.5 ^b	1.77	-52 ^{ab}	116.5
Black Racka	5	19.3 ^c	2.97	-145 ^b	46.8
Mutton Merino	5	31.2 ^a	2.15	-160 ^b	97.4
mean	30	25.3	5.20	-103	136.1

a, b, c – different superscript letters show significant differences (Tukey HSD test $p<0.05$)

As shown in Table 2, the lambs gained weight in the first grazing period (from mid-May to mid-June). The pasture-based fattening showed that the acclimatization weight loss was regained in accordance with the compensatory capacity of the young animals. The average daily gain was 199 grams (between days 106 and 138). This suggests that two and a half weeks (half of this age range) were sufficient to replace the lost weight before true growth began. Highest compensatory gain was shown by Dairy Tsigai.

Table 2: Initial weight adjusted for 106 days of age (age $p=0.254$) and subsequent daily gain (age $p=0.758$) according to the studied sheep breeds

Breeds	n	Initial weight, kg $p<0.001$	Std. Err.	Daily gain after beginning of grazing (between day 106 and 138), g $p<0.001$	Std. Err.
Tsigai	5	29.4 ^a	2.81	157 ^b	44.7
Dairy Tsigai	4	22.1 ^{bc}	3.14	307 ^a	33.4
Hung. Merino	5	23.1 ^b	1.92	240 ^{ab}	29.3
White Racka	5	24.1 ^b	1.29	144 ^c	22.9
Black Racka	5	18.0 ^c	2.58	154 ^{bc}	11.2
Mutton Merino	5	29.8 ^a	1.98	213 ^b	69.3
mean	30	24.4	4.72	199	67.2

a, b, c – different superscript letters show significant differences (Tukey HSD test $p<0.05$)

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Tables 3 and 4 illustrate that between the next two weighing periods, the lambs also lost live weight, which was also characterized by negative daily gain. These prove that the overall intervening 60 days were hindering the live weight growth. The average daily gain was -16 and -28 grams between days 138 and 165 and between days 165 and 197, respectively.

Table 3: Weight adjusted for 138 days of age (age $p=0.393$) and subsequent daily gain (age $p=0.503$) according to the studied sheep breeds

Breeds	n	Weight, kg $p<0.001$	Std. Err.	Daily gain during grazing (between day 138 and 165), g $p=0.019$	Std. Err.
Tsigai	5	34.3 ^a	3.36	14 ^{ab}	49.6
Dairy Tsigai	4	30.6 ^{ab}	3.52	-11 ^{ab}	51.0
Hung. Merino	5	30.5 ^{ab}	2.79	35 ^a	18.3
White Racka	5	28.6 ^b	1.26	-44 ^{ab}	54.5
Black Racka	5	22.8 ^c	2.65	-39 ^{ab}	18.5
Mutton Merino	5	36.4 ^a	2.87	-50 ^b	44.2
mean	29	30.5	5.15	-16	49.8

a, b, c – different superscript letters show significant differences (Tukey HSD test $p<0.05$)

In the latter phase (Table 4), we found no significant ($p=0.183$) differences between breeds, so the summer weight loss can be determined uniformly for our wether lambs under the given grazing conditions. We observed that the rate of summer weight loss was smaller than during acclimatization, so we can speak more of stagnation.

Table 4: Weight adjusted for 165 days of age (age $p=0.592$) and subsequent daily gain (age $p=0.869$) according to the studied sheep breeds

Breeds	n	Weight, kg $p<0.001$	Std. Err.	Daily gain in most detrimental period (between day 165 and 197), g $p=0.183$	Std. Err.
Tsigai	5	34.7 ^a	3.74	5	23.1
Dairy Tsigai	4	30.4 ^{ab}	2.19	-81	40.7
Hung. Merino	5	31.4 ^{ab}	2.88	-73	139.4
White Racka	5	27.4 ^{bc}	2.08	-8	17.0
Black Racka	5	21.7 ^c	2.27	4	23.5
Mutton Merino	5	35.0 ^a	3.74	-24	35.1
mean	29	30.1	5.41	-28	67.3

a, b, c – different superscript letters show significant differences (Tukey HSD test $p<0.05$)

As presented in Tables 5 and 6, the lambs also gained their weights in the last two (from mid-August to mid-October) fattening periods. The mid-summer weight losses were regained during early autumn. The average daily gain was 107 and 99 grams

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between days 197 and 228 and between days 228 and 255, respectively. In neither period did we find significant ($p=0.275$ and $p=0.352$, respectively) differences in daily growth between breeds. Thus, here too we can observe the general response of sheep. Regarding the autumn compensation, we can state that its extent is the same as that of the grazing period after acclimatization.

Table 5: Weight adjusted for 197 days of age (age $p=0.095$) and subsequent daily gain (age $p<0.009$) according to the studied sheep breeds

Breeds	n	Weight, kg $p<0.001$	Std. Err.	Daily gain after most detrimental period (between day 197 and 228), g $p=0.275$	Std. Err.
Tsigai	5	34.8 ^a	3.69	155	109.4
Dairy Tsigai	4	27.8 ^{ab}	3.42	74	17.2
Hung. Merino	5	29.1 ^{ab}	5.59	164	160.1
White Racka	5	27.1 ^{bc}	2.00	106	24.9
Black Racka	5	21.9 ^c	1.84	74	19.3
Mutton Merino	5	34.3 ^a	3.01	65	21.2
mean	29	29.2	5.56	107	85.2

a, b, c – different superscript letters show significant differences (Tukey HSD test $p<0.05$)

Table 6: Weight adjusted for 228 days of age (age $p=0.534$) and subsequent daily gain (age $p=0.089$) according to the studied sheep breeds

Breeds	n	Weight, kg $p<0.001$	Std. Err.	Daily gain during grazing (between day 228 and 255), g $p=0.352$	Std. Err.
Tsigai	5	39.6 ^a	2.96	54	124.9
Dairy Tsigai	4	30.1 ^{bc}	3.30	89	34.8
Hung. Merino	5	34.2 ^{ab}	3.66	103	30.9
White Racka	5	30.4 ^b	2.66	96	27.9
Black Racka	5	24.1 ^c	2.20	105	13.8
Mutton Merino	5	36.3 ^a	2.98	149	64.0
mean	29	32.5	5.79	99	63.6

a, b, c – different superscript letters show significant differences (Tukey HSD test $p<0.05$)

Compensatory gain, known as catch-up growth and compensatory growth, is an accelerated growth of an individual following a detrimental period of slower or even stunted development. Our study revealed that the grazing animals were affected by periods that hindered their development on two occasions: the acclimatization period following delivery and the middle of summer. At the same time, our study also revealed that the animals responded satisfactorily after both detrimental periods.

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The initial difference between breeds remained in the final weight, and this is also reflected in the average daily gain during fattening (Table 7). The animals gained on average from weight of 25.3 kg at delivery (Table 1) to a final weight of 35.2 kg. An average daily gain of 73 grams was achieved during a 149-day grazing period. This performance was largely due to castration and fattening on old-growth grassland.

In our study, castrated lambs were used in pasture-based fattening exclusively, so the results can be interpreted in this context. Castration for the purpose of extensive fattening was typically performed in the past, not only in sheep, but also in other grazers, such as goats (KOYUNCU et al., 2007) and cattle (SCHMUTZ et al., 2013; SENTURKLU et al., 2023; SZABÓ and HOLLÓ, 2026). Later, it became more typical to raise castrated lambs on pasture and then their finishing in a short intensive period (GRAVADOR et al., 2018). However, there are also examples of grazing with concentrates supplementation (AMAD et al., 2023).

Table 7: Final weight adjusted for 255 days of age (age $p=0.919$) and subsequent daily gain (age $p=0.314$) according to the studied sheep breeds

Breeds	n	Final weight, kg $p<0.001$	Std. Err.	Daily gain during the entire grazing (between day 106 and 255), g $p=0.020$	Std. Err.
Tsigai	5	41.1 ^a	3.59	79 ^{ab}	12.5
Dairy Tsigai	4	32.5 ^{bc}	2.59	77 ^{ab}	3.3
Hung. Merino	5	37.0 ^{ab}	4.23	94 ^a	16.3
White Racka	5	33.0 ^{bc}	2.53	60 ^b	15.4
Black Racka	5	27.0 ^c	2.27	61 ^b	9.2
Mutton Merino	5	40.3 ^a	3.80	71 ^{ab}	24.8
mean	29	35.2	5.85	73	18.4

a, b, c – different superscript letters show significant differences (Tukey HSD test $p<0.05$)

LINDSAY et al. (2007) found the lowest liveweight gain in pasture fattening, where the pasture was a newly established perennial ryegrass (*Lolium perenne*) and white clover (*Trifolium repens*) one. McGRATH et al. (2015) compared the growth rates and final weights of lambs grazing six novel irrigated pastures. They stated that final liveweights were highest (43.7 kg) for lambs (White Dorper) grazing the arrowleaf clover + chicory pasture.

Conclusion and recommendation

In our study, finishing lambs achieved low results in final weight and average daily gain during fattening. However, it must be admitted that under given conditions

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(castrated animals grazing) these are realistic values. During the study, weight loss occurred twice (during the acclimatization and in the middle of summer), which the animals made up for as a result of their compensatory ability and continued to grow under favourable conditions. We found that growth rates of lambs following a detrimental period are more efficient than average growth rates, in agreement with the findings of GASCOIGNE and LOVATT (2015).

Other conditions may have contributed to the poor results. For example, not only the grazing, but also the old-growth grassland itself, since the length of the grass blade here is unfavourably longer for the sheep. On the other hand, the available grass mass represents a low-to-moderate pasture cover. But we can think of the unfavourable effect of grazing without a shepherd, according to which under such conditions the animals migrate more and consume less grass.

In a more complex assessment of the economic efficiency of extensive wether fattening, other factors must also be taken into account. Traditionally, the wethers were not primarily used for meat production but were kept together with the ewes for years to shear fine wool (finer than that of rams). The wethers were intensively finished (conditioned) in late autumn to build up a strong tallow reserve for soap making in winter. Due to the close-to-nature keeping of indigenous animals, food products are highly seasonal, while other products (wool, leather, corneous material) are less so. The production and sale of the latter is therefore more balanced throughout the year. Efforts should be made to use the animals in as many ways as possible, and the income from curious - probably innovative (CILIBERTI et al., 2021) - products made from them should exceed the income from meat alone. The local animals are also an attraction for tourists with their uniqueness. Among the complementary benefits of traditional animal husbandry, we cannot forget about environmental protection. The extensive keeping of native animals is aided by the technical elements of precision livestock farming (PLF), which can be adopted from intensive farming and developed for extensive farming (HERLIN et al., 2021; SILVA et al., 2022).

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DOI: <https://doi.org/10.59913/dagr.2026.24470>

References

- AMAD, A.A. – BADEER, M. – ZENTEK, J. (2023): Effects of Feeding Systems and Castration on the Growth Performance and some Carcass Traits in Lambs of Dhamari sheep – Yemen. Arab Univ J Agric Sci, 6, 105–132. <https://doi.org/10.21608/asajs.2023.310127>
- BÖKÖNYI, S. (1974): History of Domestic Mammals in Central and Eastern Europe. Akadémiai Kiadó: Budapest, Hungary; p. 189.
- CILIBERTI, M.G. – SANTILLO, A. – MARINO, R. – CIANI, E. – CAROPRESE, M. – RILLO, L. – MATASSINO, D. – SEVI, A. – ALBENZIO, M. (2021): Lamb Meat Quality and Carcass Evaluation of Five Autochthonous Sheep Breeds: Towards Biodiversity Protection. Animals, 11, 3222. <https://doi.org/10.3390/ani11113222>
- Council Regulation (EC) No 21/2004 of 17 December 2003 establishing a system for the identification and registration of ovine and caprine animals and amending Regulation (EC) No 1782/2003 and Directives 92/102/EEC and 64/432/EEC, <http://data.europa.eu/eli/reg/2004/21/oj>
- DUNKA, B. – JÁVOR, A. (1991): Maintaining the two colour variants of the Racka through line breeding (in Hungarian). In Őshonos és honosult háziállatfajtáink genetikai sajátosságai II. Kutatási jelentések; Bodó, I. ed.; Budapest; pp. 28–36.
- DUNKA, B. (2006): The Hungarian Sheep (*Ovis aries strepsiceros hungaricus*) (in Hungarian); In Old Hungarian sheep breeds; Jávör, A.; Kukovics, S. Eds.; Mezőgazda Kiadó: Budapest, Hungary; pp. 13–30.
- FÉSÜS, L. (1991): New results of immunogenetic studies of our indigenous sheep breeds (in Hungarian). In Őshonos és honosult háziállatfajtáink genetikai sajátosságai II. Kutatási jelentések; Bodó, I. ed.; Budapest; pp. 37–48.
- GAÁL, L. (1986): The history of Hungarian animal husbandry (in Hungarian). Akadémiai Kiadó: Budapest, Hungary; pp. 43–46.
- GASCOIGNE, E. – LOVATT, F. (2015): Lamb growth rates and optimising production. Farm Animals, 37, 401–417.
- GÁSPÁRDY, A. – SIMON, Cs. – ANDRÁSOF SZKY, E. – SÁFÁR, L. – KÓSA, E. (2016): Historical Evaluation of milk producing ability of the Hungarian native Tsigai sheep (in Hungarian). Állattenyésztés és Takarmányozás, 65, 24–36.
- GRAVADOR, R.S. – MOLONEY, A.P. – BRUNTON, N.P. – GKARANE, V. – ALLEN, P. – FAHEY, A.G. – CLAFFEY, N.A. – DISKIN, M.G. – FARMER, L.J. – MONAHAN, F.J. (2018): Effects of castration and slaughter age on the fatty acid composition of ovine muscle and adipose tissue from two breeds. Small Rumin Res, 168, 94–100. <https://doi.org/10.1016/j.smallrumres.2018.09.018>.
- HANKÓ, B. (1937): The origin, past and present of the Hungarian sheep (in Hungarian). Special edition from Volume 2 of “TISIA”, Nagy Könyvkiadó: Debrecen, Hungary

DOI: <https://doi.org/10.59913/dagr.2026.24470>

- HERLIN, A. – BRUNBERG, E. – HULTGREN, J. – HÖGBERG, N. – RYDBERG, A. – SKARIN, A. (2021): Animal Welfare Implications of Digital Tools for Monitoring and Management of Cattle and Sheep on Pasture. *Animals*, 11, 829. <https://doi.org/10.3390/ani11030829>
- KOYUNCU, M. – DURU, S. – KARA UZUN, Ş. – ÖZİŞ, Ş.; – TUNCEL, E. (2007): Effect of castration on growth and carcass traits in hair goat kids under a semi-intensive system in the south-Marmara region of Turkey, *Small Rumin Res*, 72, 38–44, <https://doi.org/10.1016/j.smallrumres.2006.08.001>
- LINDSAY, C.L. – KEMP, P.D. – KENYON, P.R. – MORRIS, S.T. (2007): Summer lamb finishing on forage crops. *Proceedings of the New Zealand Society of Animal Production*, 67, 121–125.
- MATOLCSI, J. (1975): The origin of domestic animals (in Hungarian). *Mezőgazdasági Kiadó: Budapest, Hungary*, pp. 149–159.
- McGRATH, S. – SANDRAL, G. – FRIEND, M. (2015): Lamb growth rates on pasture: assessing options for finishing lambs in spring. © 2015 “Building Productive, Diverse and Sustainable Landscapes”, *Proceedings of the 17th ASA Conference*, 20 – 24 September 2015, Hobart, Australia. Web site <http://www.agronomy2015.com.au>
- SENTURKLU, S. – LANDBLOM, D. – STOKKA, G. – CIHACEK, L. (2023): Perspective Chapter: Alternative Intensive Animal Farming Tactics That Minimize Negative Animal Impact and Improve Profitability. In *Intensive Animal Farming - A Cost-Effective Tactic*; Manzoor, S.; Abubakar, M. Eds.; IntechOpen; <https://doi.org/10.5772/intechopen.108339>
- SILVA, S.R. – SACARRÃO-BIRRENTTO, L. – ALMEIDA, M. – RIBEIRO, D.M. – GUEDES, C. – GONZÁLEZ MONTAÑA, J.R. – PEREIRA, A.F. – ZARALIS, K. – GERALDO, A. – TZAMALOUKAS, O. – CABRERA, M.G. – CASTRO, N. – ARGÜELLO, A. – HERNÁNDEZ-CASTELLANO, L.E. – ALONSO-DIEZ, Á.J. – MARTIN, M.J. – CAL-PEREIRA, L.G. – STILWELL, G. – DE ALMEIDA, A.M. (2022): Extensive Sheep and Goat Production: The Role of Novel Technologies towards Sustainability and Animal Welfare. *Animals*, 12, 885. <https://doi.org/10.3390/ani12070885>
- SCHANDL, J. (1955): Sheep farming (in Hungarian). *Mezőgazdasági Kiadó: Budapest, Hungary*; pp. 78–80.
- SCHMUTZ, M. – WEINDL, P. – CARRASCO, S. – BELLOF, G. – SCHMIDT, E. (2013): Effect of breed, grazing system and concentrate supplementation on fattening performance, carcass value and meat quality of steers. *Archiv Tierzucht*, 56, 943–957. <https://doi.org/10.7482/0003-9438-56-096>
- SZABÓ, F. – HOLLÓ, G. (2026): The Necessity and Feasibility of Implementing Regenerative Practices in Pasture-Based Beef Cattle Farming. *Animals*, 16(15), 2384. <https://doi.org/10.3390/ani16152384>

DOI: <https://doi.org/10.59913/dagr.2026.24470>

SZABÓ, M. – KUSZA, Sz. – CSÍZI, I. – MONORI, I. (2016): The place of the Hungarian Merino within the Merino breed group (in Hungarian). *Agrártudományi Közlemények*, 69, 151–156.

TIBCO Software Inc. Data Science Workbench. Statistica Version 14. 2020.

DOI: <https://doi.org/10.59913/dagr.2026.23866>

Conservation and Management of the Carniolan Honey Bee (*Apis mellifera carnica*) in Slovenia

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Abstract

The Carniolan honey bee (*Apis mellifera carnica*) is an indigenous Slovenian subspecies with a long tradition and considerable importance for agriculture and ecosystem functioning. In Slovenia, it is legally protected, as only this subspecies may be bred, ensuring the preservation of its genetic integrity. It is characterized by gentleness, high productivity and adaptability, traits that have contributed to its widespread use worldwide. Beekeeping is an important agricultural activity in Slovenia; more than 12,000 beekeepers manage approximately 210,000 bee colonies, evenly distributed across the country. The number of colonies and queens has increased in recent years, suggesting positive population trends and potential effectiveness of breeding and conservation measures. Beyond the production of honey and other bee products, the principal contribution of honey bees is pollination, a vital ecosystem service that supports biodiversity and agricultural productivity. Consequently, the conservation of the Carniolan honey bee is essential for sustainable agriculture, ecosystem stability, and food security.

This article examines the status, population trends, and conservation of the Carniolan honey bee (*Apis mellifera carnica*) in Slovenia based on national statistical data and registry records. Particular attention is given to its spatial distribution, breeding practices, and honey production dynamics during the period 2010–2025.

Keywords: Carniolan honey bee (*Apis mellifera carnica*); beekeeping in Slovenia; honey production; pollination; ecosystem services

Introduction

Bees are among the most important animal pollinators and play a crucial role in agricultural production, biodiversity conservation, and ecosystem functioning. It is estimated that approximately one-third of global food production depends directly or indirectly on animal-mediated pollination, with honey bees constituting one of the most significant pollinator groups. In addition to supporting crop yields and quality, pollination services are essential for the reproduction of many wild plant species and the maintenance of ecosystem stability. The economic value of pollination substantially exceeds the market value of all bee-derived products combined, underscoring the critical importance of bees for both natural and agricultural systems. Slovenia is recognised as one of the leading beekeeping countries in Europe, characterised by a rich and long-standing apicultural tradition. Historical records indicate that beekeeping was already well established in the Slovenian territory by the seventeenth century, as documented by Janez Vajkard Valvasor in *The Glory of the Duchy of Carniola* (1689). The scientific significance of Slovenian beekeeping was further strengthened in 1770, when Giovanni Antonio Scopoli described the Carniolan honey bee in his work *Dissertatio de Apibus*. The currently accepted taxonomic designation, *Apis mellifera carnica*, was subsequently established by Pollmann in 1879. The Carniolan honey bee (*Apis mellifera carnica*) is one of Slovenia's most important indigenous animal genetic resources and represents a distinctive component of the country's natural and cultural heritage. Owing to its gentleness, strong foraging behaviour, excellent orientation abilities, and adaptability to diverse environmental conditions, it is widely regarded as one of the most valuable subspecies of the western honey bee (*Apis mellifera*). These characteristics contributed to its extensive dissemination throughout Europe during the nineteenth century and later to other parts of the world. Recognising its ecological, economic, and cultural significance, Slovenia has granted the Carniolan honey bee special legal protection and designated it as the only honey bee subspecies permitted for breeding and management within the country. Nevertheless, despite comprehensive conservation and breeding efforts, the subspecies is increasingly exposed to multiple environmental and biological pressures, including infestations by *Varroa destructor*, climate change, fluctuations in nectar and pollen availability, habitat degradation, and agricultural intensification. These factors may adversely affect colony health, productivity, and the long-term preservation of genetic diversity. Against this background, the present paper examines the role and significance of the Carniolan honey bee in Slovenia. Particular attention is given to the characteristics of its breeding and conservation programmes, population trends, beekeeping activities, and honey production. The paper also evaluates the contribution of the Carniolan honey bee to pollination services, biodiversity conservation, and food security, while discussing the principal challenges affecting its long-term sustainability and management.

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Characteristics, Legal Protection, and Breeding Management of the Carniolan Honey Bee

The Carniolan honey bee (*Apis mellifera carnica*) is distinguished by the characteristic greyish hairs covering its thorax and is widely recognised as one of the most valuable subspecies of the western honey bee (*Apis mellifera*). Its principal characteristics include a gentle temperament, excellent orientation ability, rapid spring population development, efficient utilisation of available forage resources, strong adaptability to diverse environmental conditions, and high honey production potential. Owing to this combination of favourable traits, the Carniolan honey bee has gained international recognition and has been widely disseminated beyond its native range (Gregori, 2012). The introduction of non-native honey bee subspecies has led to the decline or loss of indigenous honey bee populations in many parts of the world. In Slovenia, however, the native Carniolan honey bee has been successfully preserved through long-term conservation efforts. The first formal initiative aimed at protecting its genetic integrity dates back to 1920, when the Memorandum of the Museum Society for Slovenia highlighted the importance of maintaining the genetic purity of the Carniolan honey bee (Skoberne, 2020). Conservation of the subspecies has subsequently been reinforced through national legislation and international commitments. Slovenia's accession to the European Union further strengthened the institutional framework for its protection, while the Resolution on the Protection of the Carniolan Honey bee reaffirmed the country's commitment to implementing measures necessary for its long-term preservation. Today, the Carniolan honey bee is protected under the Livestock Breeding Act, which recognises it as an indigenous breed of special national importance. Accordingly, the breeding and management of honey bees in Slovenia are restricted exclusively to the Carniolan honey bee, and the introduction of other honey bee subspecies is prohibited (Livestock Breeding Act, Official Gazette of the Republic of Slovenia No. 18/2002, as amended). A central instrument for the conservation and sustainable development of the Carniolan honey bee population is the Breeding Programme for the Carniolan honey bee (Kozmus et al., 2023). The programme aims to preserve genetic integrity and diversity, maintain a viable population size, improve economically important traits, and ensure the production of high-quality breeding queens. Since 2004, a studbook (pedigree register) has been maintained as a key tool for monitoring pedigrees, evaluating breeding stock, and managing selection activities. The breeding programme is implemented through cooperation between the recognised breeding organisation, beekeepers, research institutions, and governmental authorities, thereby ensuring the long-term conservation, genetic improvement, and sustainable management of the Carniolan honey bee population in Slovenia (Figure 1 and 2).



Figure 1: Queen bee on a honeycomb surrounded by worker bees (Photo: Tratnjek)



Figure 2: Slovenia is home to more than 10,000 architecturally distinctive apiaries, a unique feature not found elsewhere in the world, reflecting the country's rich beekeeping tradition and cultural heritage (Photo: ČZS)

Material and methods

Data on honey bee colonies, apiaries, beekeepers, queen production, pedigree queens, honey production, and honey self-sufficiency were obtained from the Central Register of Apiaries and Beekeepers maintained by the Administration of the Republic of Slovenia for Food Safety, Veterinary Sector and Plant Protection. Data on queen production and pedigree queens were obtained from the Agricultural Institute of Slovenia. Honey production and self-sufficiency data were retrieved from the Statistical Office of the Republic of Slovenia (SURs). Descriptive statistics and graphical analyses were performed using Microsoft Excel.

Results and discussion

The Carniolan honey bee population in Slovenia

According to the Central Register of Apiaries and Beekeepers, Slovenia has more than 12,000 registered beekeepers, each managing an average of 18.7 honey bee colonies. The predominant hive type is the Albert–Žnideršič hive (AŽ, 2020), which accommodates approximately 93% of all managed colonies in the country. Under the Rules on bee houses and bee yard identification (Official Gazette of the Republic of Slovenia, No. 117/2008), beekeepers are required to report the number of colonies they manage to the Central Apiary Register twice annually, on 15 April and 31 October. During the period 2015–2024, the average number of honey bee colonies recorded in Slovenia was 191,548 (Agricultural Institute of Slovenia, 2025). Honey bee colonies are distributed throughout the national territory; however, their spatial density exhibits considerable geographic variation. Higher colony densities are

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typically observed in valleys and foothill regions, whereas lower densities occur in remote and extensively forested areas. With an average density of approximately 7.5 colonies per square kilometre, Slovenia ranks among the European countries with the highest densities of managed honey bee colonies (ČZS, 2025). Maintaining a sufficiently large and spatially well-distributed population of honey bee colonies therefore represents one of the principal objectives of the Resolution on the Protection of the Carniolan honey bee (*Apis mellifera carnica*) (Official Gazette of the Republic of Slovenia, No. 18/2014, as amended).

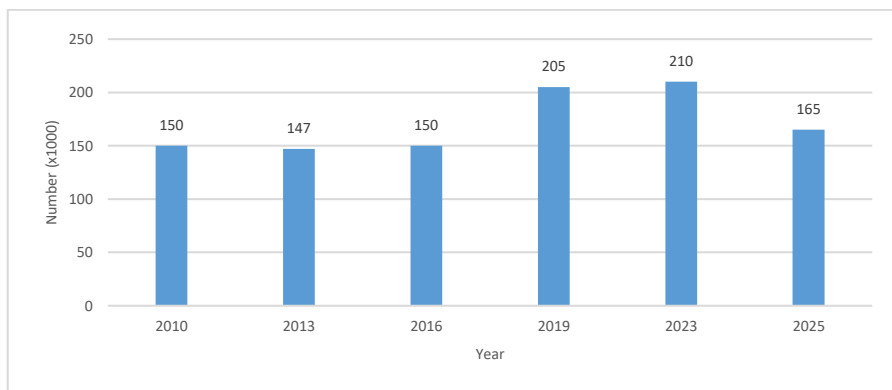


Figure 3. Number of Carniolan honey bee colonies in Slovenia, 2010–2025 (Register ..., 2025).

Figure 3 presents trend in the number of Carniolan honey bee colonies in Slovenia between 2010 and 2025. The data reveal a clear long-term upward trend, with colony numbers increasing from approximately 150,000 in 2010 to more than 210,000 in 2023. This increase reflects the strong tradition of beekeeping in Slovenia, the successful conservation and management of the Carniolan honey bee, and the continued interest of beekeepers in maintaining and expanding their apiaries. The fitted trend line further corroborates the overall growth of the population throughout the study period. A pronounced decline is apparent in the 2025 data. However, this decrease should not be interpreted as evidence of an actual reduction in the Carniolan honey bee population. Rather, it is primarily attributable to a lower reporting rate to the Central Apiary Register (Administration..., 2025). As substantially fewer beekeepers submitted colony records in 2025 compared with previous years, the reported number of colonies is likely underestimated and therefore not directly comparable with earlier observations. When this reporting-related artefact is taken into account, the data indicate that the Carniolan honey bee population in Slovenia has remained stable and has generally exhibited positive growth over the past decade.

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Queen breeding and registered pedigree queens of the Carniolan honey bee in Slovenia

Queen breeding constitutes a fundamental component of modern apiculture and plays a crucial role in preserving the desirable phenotypic characteristics and genetic integrity of the Carniolan honey bee (*Apis mellifera carnica*). In Slovenia, queen breeding is conducted at officially approved breeding stations operating within the framework of the national breeding programme. At present, 36 registered breeding stations are actively involved in queen production.

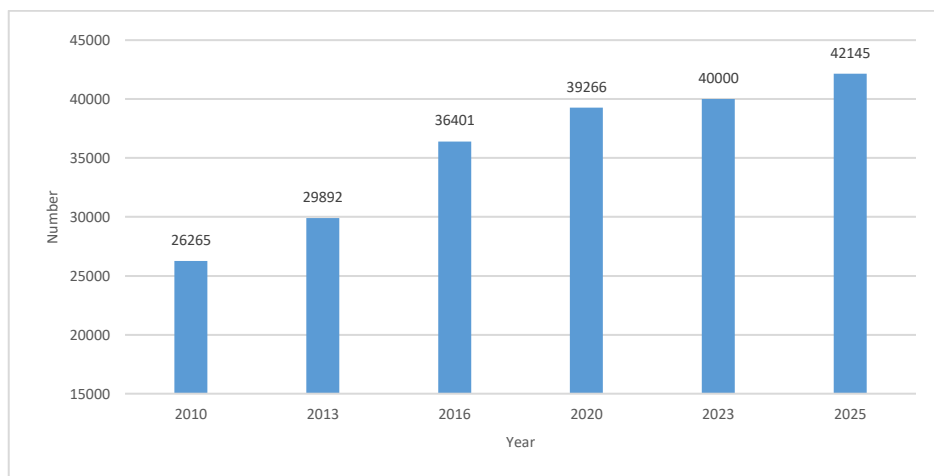


Figure 4. Trends in the production of Carniolan honey bee queens in Slovenia, 2010–2025 (Agricultural..., 2026)

Figure 4 illustrates temporal trends in the production of Carniolan honey bee queens in Slovenia between 2010 and 2025. The data reveal a pronounced long-term increase in queen production, despite several short-term fluctuations. Following a production peak in 2019, the number of queens produced temporarily declined before recovering in subsequent years. Nevertheless, the overall positive trend indicates the sustained success of organised queen-rearing activities and underscores the importance of structured breeding programmes for the conservation, genetic improvement, and long-term sustainability of the Carniolan honey bee population in Slovenia. Registered pedigree queens constitute the cornerstone of both genetic improvement and conservation efforts. Their production is based on the rigorous selection of breeder colonies exhibiting desirable morphometric, behavioural, and production-related characteristics. The selection process is coordinated and supervised by the Slovenian Beekeeper's Association (ČZS, 2025) in cooperation with the Agricultural Institute of Slovenia (Agricultural..., 2025)

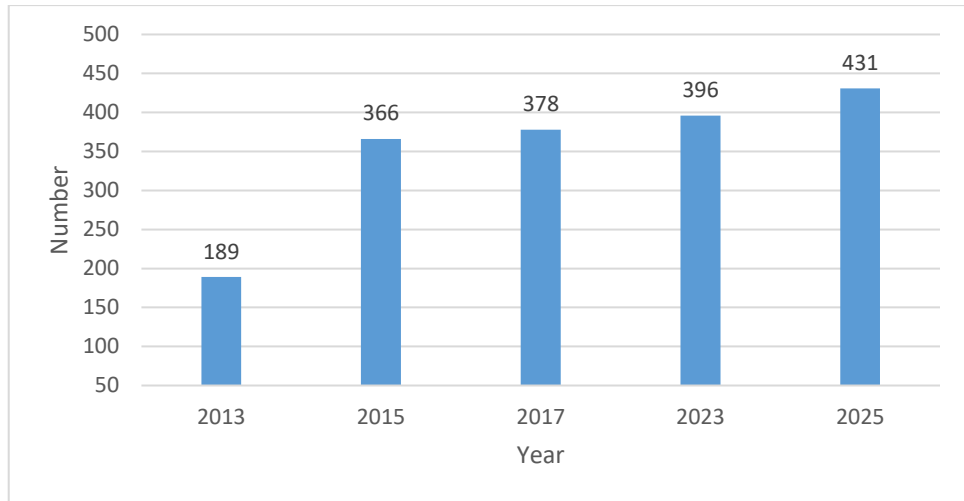


Figure 5. Number of registered pedigree queens of the Carniolan honey bee in Slovenia, 2013–2025 (Agricultural..., 2026)

Figure 5 presents the number of registered pedigree queens between 2013 and 2025. Although substantial interannual variation was observed throughout the study period, the overall trend indicates a gradual increase in production, culminating in 2025. This development reflects the continued strengthening of organised breeding activities and the growing emphasis placed on the conservation and genetic improvement of the Carniolan honey bee.

Number of bee keepers in Slovenia

Slovenia has a long-standing and well-developed beekeeping tradition, reflected in the continuous growth of its beekeeping community. In contrast to the overall declining trend observed across the European Union, the number of Slovenian beekeepers has increased by approximately 30% in recent years, accounting for around 1.1% of all beekeepers in the EU (World Bee Day, 2018). On average, Slovenian beekeepers manage approximately 16 honey bee colonies, although a smaller proportion of professional and semi-professional beekeepers operate considerably larger apiaries.

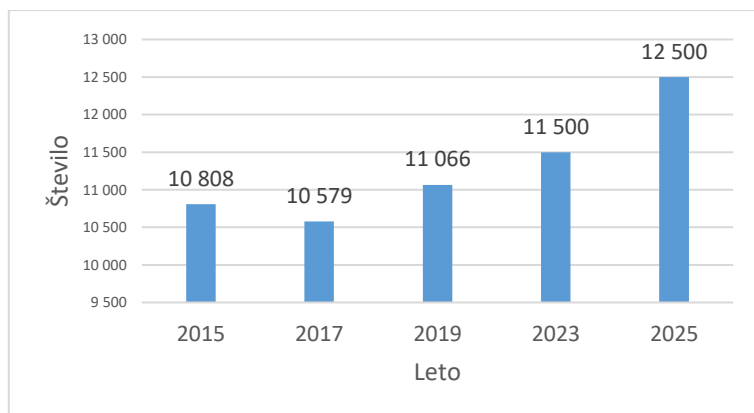


Figure 6. Number of beekeepers in Slovenia, 2015–2025 (Agricultural..., 2026)

Figure 6 illustrates trends in the number of registered beekeepers in Slovenia between 2015 and 2025. Throughout the study period, the number of registered beekeepers exceeded 12,000, corresponding to approximately five to six beekeepers per 1,000 inhabitants. The data reveal a consistent upward trend, indicating sustained interest in beekeeping and reaffirming the importance of apiculture within Slovenian society. The observed increase in beekeeper numbers may be attributed to a combination of favourable institutional support, targeted educational and promotional activities, and growing public awareness of the ecological and economic importance of pollinators. More broadly, these trends underscore the continuing role of beekeeping not only as an agricultural activity but also as a significant cultural and socio-economic component of Slovenia's rural landscape and national heritage.

Geographical distribution of the Carniolan honey bee in Slovenia

During the period 2015–2024, an average of 191,548 honey bee colonies and approximately 14,000 apiaries were registered in Slovenia, distributed throughout the country.

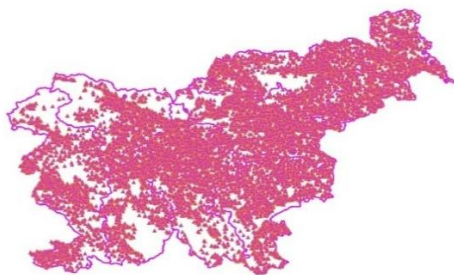


Figure 7. Geographical distribution of apiaries with the Carniolan honey bee in Slovenia (Administration..., 2025)

Figure 7 presents the spatial distribution of registered apiaries housing the Carniolan honey bee in Slovenia. Individual points represent apiary locations recorded in the national register and reveal a dense and widespread distribution across the national territory. Apiaries are present in all regions of the country, indicating that beekeeping is a well-established and geographically extensive agricultural activity. The highest concentrations of apiaries occur in lowland, hilly, and agriculturally productive areas, particularly in central, eastern, and southeastern Slovenia. In contrast, lower apiary densities are evident in the Alpine region and in sparsely populated mountainous and heavily forested areas, where environmental conditions, land use patterns, and accessibility may be less favourable for beekeeping activities. The observed spatial pattern reflects both the long-standing apicultural tradition of Slovenia and the suitability of much of the country for honey bee management. Furthermore, the broad geographical distribution of apiaries contributes to the maintenance of a well-dispersed population of the Carniolan honey bee, thereby supporting pollination services, genetic connectivity among populations, and the long-term conservation of this indigenous subspecies. Overall, the near-nationwide distribution of apiaries highlights the ecological, agricultural, and socio-cultural importance of beekeeping within Slovenia.

Honey Production 2010–2025

Slovenian beekeepers produce honey protected by a Protected Geographical Indication (PGI), which certifies both its geographical origin and compliance with defined quality standards. Slovenian honey obtained national protection in 2008 and was subsequently granted European Union protection in 2013. Producers participating in the PGI scheme are subject to stringent quality control procedures, and honey marketed under the protected label must comply with relevant national and European legislation governing honey quality, authenticity, and traceability (ČZS, 2025). Honey consumption in Slovenia remains relatively stable and averages approximately 1.4 kg per capita annually.

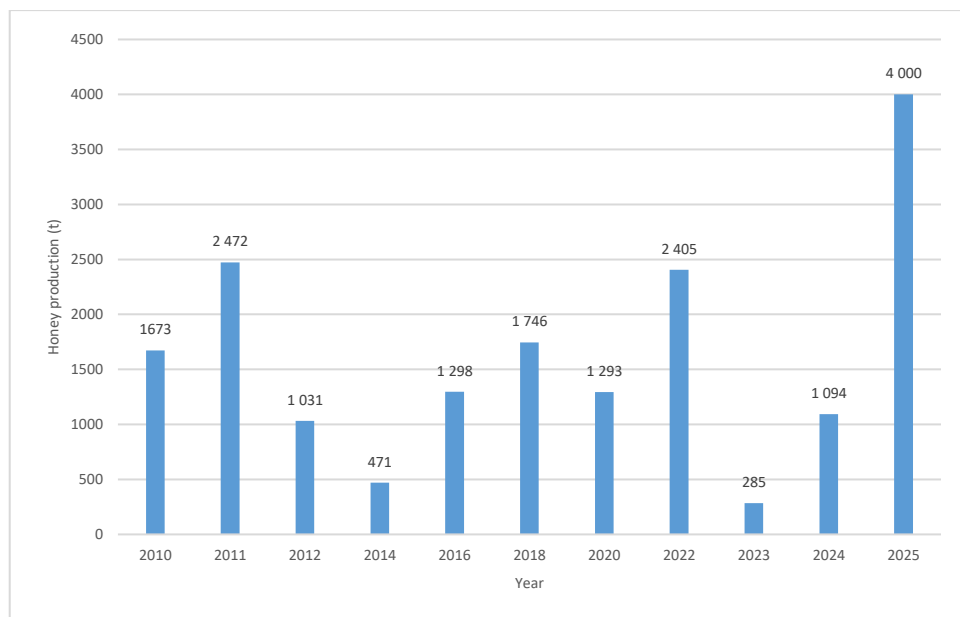


Figure 8. Honey Production in Slovenia over the period 2010–2025 (SURS, 2026)

Figure 8 presents annual honey production in Slovenia between 2010 and 2025. The results reveal considerable interannual variability, reflecting the strong influence of climatic and environmental conditions on honey yields. The highest production during the observed period was recorded in 2011 (2,472 tonnes), while the lowest occurred in 2023 (285 tonnes). Honey production was also exceptionally high in 2022, approaching the record level achieved in 2011. Following the poor harvest in 2023, production recovered substantially in 2024. According to provisional estimates of the Statistical Office of the Republic of Slovenia (SURS, 2026), honey production reached approximately 4,000 tonnes in 2025, representing one of the highest yields recorded in recent decades. This exceptional harvest was largely attributed to favourable weather conditions, abundant nectar flows, and extended foraging opportunities. However, the data for 2025 are preliminary and should be interpreted with caution. Overall, the results demonstrate that honey production in Slovenia is highly dependent on environmental conditions. Despite relatively stable colony numbers and continued growth of the beekeeping sector, annual honey yields remain strongly influenced by climatic variability and nectar availability.

Honey self-sufficiency in Slovenia, 2005–2025

Honey consumption in Slovenia has remained relatively stable over the past two decades, averaging slightly above 1 kg per capita annually. In contrast, the honey self-sufficiency rate has shown marked interannual variability, primarily driven by

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fluctuations in domestic honey production linked to climatic and environmental conditions. Periods of favourable weather and abundant nectar availability have supported high levels of self-sufficiency, whereas adverse conditions have led to reduced domestic yields and increased dependence on imports. This was particularly evident in 2014 and 2021, when domestic production did not meet national demand. In subsequent years, honey production recovered significantly, most notably in 2022 and again in 2025, resulting in a substantial improvement in the self-sufficiency rate. The exceptionally high level recorded in 2025 indicates that domestic production exceeded national consumption requirements, generating a production surplus. Overall, these patterns highlight the strong dependence of Slovenia's honey sector on annual harvest outcomes and underscore the dominant influence of climatic variability on the country's ability to meet domestic demand through local production.

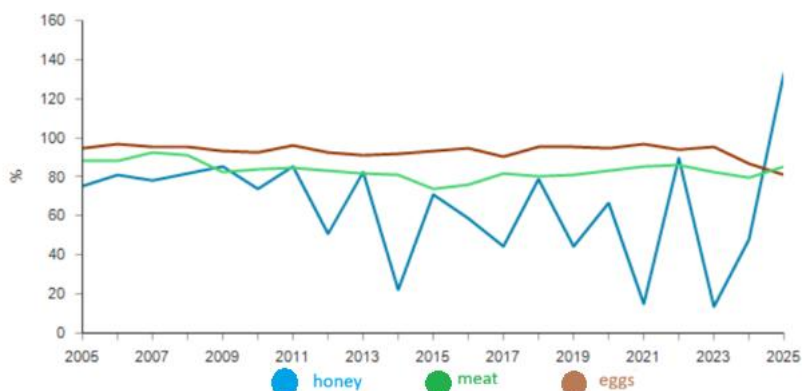


Figure 9. Self-sufficiency rate in honey production in Slovenia, 2005–2025 (SURS, 2026)

Figure 9 compares self-sufficiency rates for honey, meat, and eggs in Slovenia between 2005 and 2025. While meat and egg self-sufficiency remain relatively stable over time, honey shows considerably greater fluctuations. These variations reflect the strong dependence of honey production on weather conditions, nectar availability, and other environmental factors. Overall, the graph highlights the high variability of honey self-sufficiency compared to the much more consistent trends observed for other livestock products.

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Conclusion and recommendation

The Carniolan honey bee (*Apis mellifera carnica*) represents one of Slovenia's most valuable indigenous animal genetic resources and a cornerstone of the country's long-standing beekeeping tradition. The results of this study indicate a generally positive development of the Slovenian beekeeping sector over the past decade, reflected in increases in the number of honey bee colonies, registered beekeepers, and pedigree queen production. These trends demonstrate the effectiveness of existing conservation and breeding programmes and confirm the continued vitality of beekeeping in Slovenia. The widespread spatial distribution of apiaries across the country further highlights the extensive geographical presence of beekeeping and contributes to a well-dispersed honey bee population. At the same time, the results reveal substantial interannual variability in honey production and honey self-sufficiency, primarily driven by climatic and environmental conditions that affect nectar availability and foraging opportunities. This highlights the sensitivity of honey production to annual weather patterns, despite the relative stability of colony numbers. The exceptionally high honey yield recorded in 2025 illustrates the production potential of the sector under favourable conditions, whereas poor harvest years demonstrate its vulnerability to adverse climatic events. Future sustainability of the Slovenian beekeeping sector will therefore depend not only on continued conservation and breeding efforts, but also on adaptation to increasing environmental variability associated with climate change. Beyond its economic role, the Carniolan honey bee provides essential pollination services that support agriculture, biodiversity, ecosystem functioning, and food security. Ensuring the long-term preservation of its genetic integrity, maintaining a sufficiently large and well-distributed population, and improving forage availability and environmental resilience remain key priorities for its sustainable management.

References

- ADMINISTRATION OF THE REPUBLIC OF SLOVENIA FOR FOOD SAFETY, VETERINARY SECTOR AND PLANT PROTECTION. (2025). (<https://www.gov.si/drzavni-organi/organi-v-sestavu/uprava-za-varno-hrano-veterinarstvo-in-varstvo-rastlin/o-upravi/>)
- AGRICULTURAL INSTITUTE OF SLOVENIA. (2025). Sustainable Food Supply in Slovenia. (<https://eionet.arso.gov.si/>)
- AGRICULTURAL INSTITUTE OF SLOVENIA. (2026). (<https://www.kis.si/>)
- AŽ hive, 2020. ČZS - <https://www.czs.si/content/D11>
- ČEBELARSKA ZVEZA SLOVENIJE (ČZS). (2025). <http://www.czs.si/>
- GREGORI, J. 2012. Kranjska čebela. Zloženka

DOI: <https://doi.org/10.59913/dagr.2026.23866>

KOZMUS, P. – PODGORŠEK, P. – BOŽIČ, J. – ŠMERC, F. – JAVORNIK, F. – GREGORI, J. – HRASTELJ, M. – DREMELJ, J. 2023. Breeding program for Carniolan Honey bee 2018-2023 (*Apis mellifera carnica*, Pollmann 1879). (http://www.czs.si/Files/Rejski%20program%20za%20KRANJSKO%20CEBELO%20_2018-2023.pdf)

LIVESTOCK BREEDING ACT, Official Gazette of the Republic of Slovenia No. 18/2002

POLLMANN, A. (1879). Original taxonomic description of *Apis mellifera carnica*. REGISTER OF BREEDS WITH ZOOTECHNICAL ASSESSMENT. (2025). (<http://www.genskabanka.si/wcontent/uploads/2025/02/RegisterPasem2025.xls>)

RESOLUTION ON THE PROTECTION OF THE CARNIOLAN HONEY BEE (*Apis mellifera carnica*, Pollmann 1879) (2014). Official Gazette of the Republic of Slovenia No. 18/14

RULES ON THE MARKING OF APIARIES AND BEEHIVE SITES. (2008): Official Gazette of the Republic of Slovenia, No. 117/2008, 12 December 2008

SCOPOLI, G.A. (1770): *Dissertatio de Apibus*. In: *Annus IV. Historico-Naturalis*, Hilscherus, Lipsiae (Leipzig). pp. 7–47.

SKOBERNE, P. (2020): *Spomenica 1920 - prvi nacionalni program za varstvo narave v Sloveniji*. *Trdoživ*, 9, 1: 16-20, URN:NBN:SI:DOC-Y1SUOYE7 from <http://www.dlib.si>

SURS. (2026). <http://www.stat.si/statweb>

SURS. (2025). <https://www.stat.si/statweb/News/Index/11125>

VALVASOR, J. W. (1689). *Die Ehre des Herzogthums Krain* (The Glory of the Duchy of Carniola)

WORLD BEE DAY. (2018). (<https://www.stat.si/StatWeb/File/DocSysFile/10430/sl-dan-cebel.pdf>)