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# Danubian Animal Genetic Resources

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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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DOI: <https://doi.org/10.59913/dagr.2024.18112>

## **Research on the European wild boar population (*Sus scrofa crofa L.*) in the Timiș county, Romania**

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### **Abstract**

The wild boar represents one of the most required species by the hunters of Romania and abroad. Thus, in this paper, the evolution of wild boars population in the Timiș county (the west of Romania) is presented for a period of 10 years, from 2014 to 2023. During the year 2019, the highest number of specimens had been registered, namely, 3122 and in the following years the livestock decreased mainly because of the African swine fever (ASF). To get a management of the best quality, concerning the hunting economics and hunting mainly, there is the Hunting Complex in Sarlota, possessing an enclosed terrain of 1204 ha, where the wild boar represents one of the main species. There is a hunting museum nearby, that is a good source of professional information. In the Barzava area, in the Arad county, hybridization is practised rarely for commercial use, between the domestic swine and the wild boar.

Keywords: wild boar, game, hunting park

### **Introduction**

The increasing request for game meat in restaurants make its processing already an industry. The European wild boar (*Sus scrofa scrofa L.*) represents one of the most widespread game species in Romania, being found from the Danube Meadow to the mountains. This “senior of the forest” as ROUCAUTE (1973) called it prefers the forest as a biotope but also areas covered in rich grass “this being proof of its ecological plasticity” CRACIUNESCU (2014).

Sometimes this species is criticised for the damages it produces to agriculture, albeit debatable.

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Figure 1. Wild boar from Timis Country. Orig. Matiuti M.

## Material and methods

The data in the scientific paper come from the Ministry of Forests and Water as well as from the Transylvanian Rare Breeds Association's (TRB) database for the period 2014-2023. These data refer to the wild boar population in the Timiș country, in the West of Romania (Figure 1).

## Results and discussion

According to the literature in the field (MEYNHARD, 1988; SELARIU, 1995) wild boar evaluation is done annually, the methods used to be mainly the direct observation and tracks reading in the spring after the last snowfall. „This is because after winter has passed mortality stops in wild boar herds” (SELARIU, 2001).

Figure 2 presents the evolution of wild boar herds in Timiș County in the period 2014-2023.

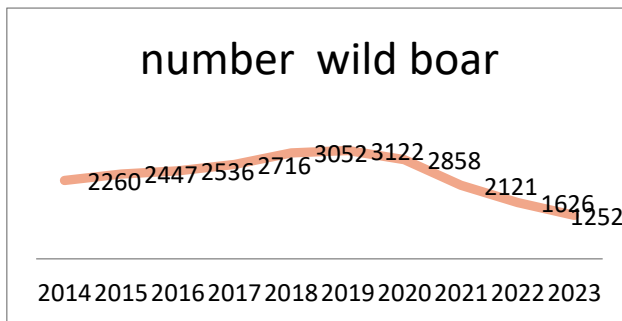


Figure 2. The evolution of wild boar herds in the period 2014-2023 (according to the Ministry of Environment, Water and Forests, 2023)



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The diagram shows an increase in wild boar herds in 2019, when they reached 3122 specimens, then following an accelerated decrease in their number. The increase is due to high temperatures during winter and food abundance, followed by a decrease mainly caused by the African swine fever (ASF) starting in 2021 (UNGUR et al., 2021).

For a qualitative management of the game economy and hunting in Timiș County, there is the Sarlota Hunting Complex which operates in a deciduous forest with a fenced area of 1204 ha under the administration of the National Forestry Administration (Figure 3). This is a continuation of the Hunting Park established in 1904 by the Austrian count Siegfried von Wimpffen for two important reasons: "The organization of hunting parties and the protection of agricultural crops on the estates around the Sarlota forest, against the damage caused by wild animals" (CRACIUNESCU and GARGAREA, 2017).

The natural feeding conditions, as well as the complementary feeding offer very good conditions for the main species found here: the European fallow deer (*Dama dama*), the wild boar and the European mouflon (*Ovis aries musimon P.*).

In the park there are hunting installations and facilities, observatories, feeders, salt pans, hay storages, water wells.

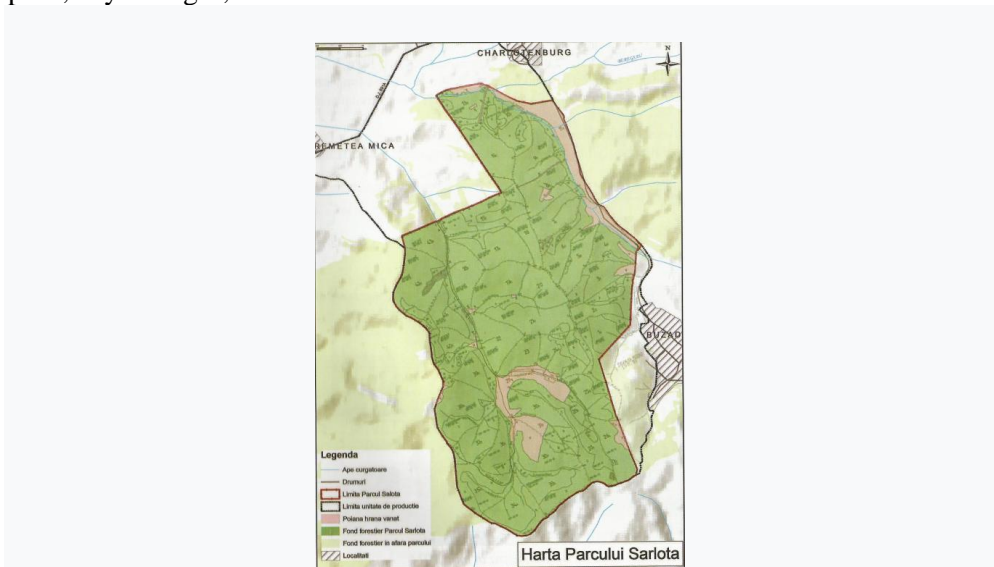


Figure 3. Map of the Sharlota Hunting Park (after CRACIUNESCU and GARGAREA 2017)

In 2014, the Charlottenburg Hunting Museum was inaugurated near the park, which represents a source of information on the evolution of game and hunting in Banat,

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with a rich collection of trophies, taxidermy and photographs from different periods of the existence of the hunting park (Figure 4).



Figure 4. left: Hunting Museum. Orig. Matiuti Carmen, right: Hybrid from Barzava. Database TRB

For commercial purposes rarely in some areas of western Romania, for example Barzava (Arad county), hybridization between domestic pig and wild boar is being practiced (Figure 4).

The resulting products have the boar-like phenotype and are sold to private hunting funds. This practice is almost abandoned today. The research undertaken in the Barzava area during 2002-2010 demonstrate this according to Figure 5 (MATIUTI et al., 2010).

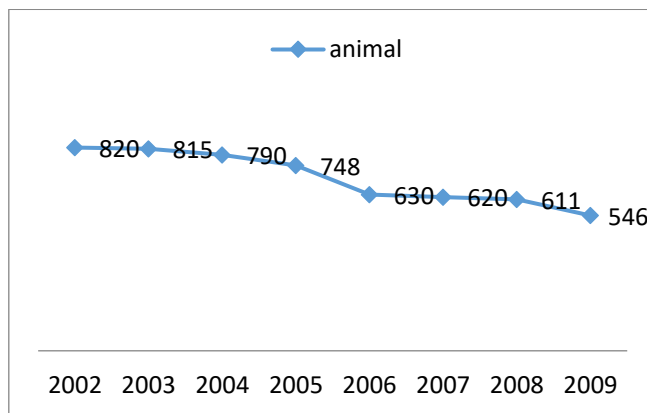


Figure 5. Number of hybrids in period 2002-2009

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## Conclusion and recommendation

The wild boar remains a valuable species for trophies, meat, fur and heart valves which are used in heart surgery. The healthy and vigorous wild boar population from Romania won numerous gold, silver and bronze medals in international hunting trophies contests.

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## Maternal genetic diversity of the Red-faced sheep of Covasna based on the mtDNA control region

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### Abstract

One of the variants of the Tsigai sheep is the reddish-headed and -legged, small-sized, so-called Redface Covasna. In 2014, the Institute for Farm Animal Gene Preservation brought in a small herd of these from Transylvania with the aim of national conservation of the breed. The aim of the study is to characterize the maternal background and maternal genetic diversity of the Red-faced sheep of Covasna. For this, the nucleotide sequence of the control region (CR) of the mitochondrial genome (mtDNA) was used. The sampling was achieved from 34 individuals 2021 and 2022. The length of the aligned CR sequences is 621 bp with the number of polymorphic base sites 88. The number of haplotypes is 30 (from haplogroups A and B, which proves its origin in Asia Minor). The value of haplotype diversity ( $H_d$ ) is 0.991, and its variance is 0.010. The mean of the nucleotide diversity ( $\pi$ ) and the average nucleotide difference per pair ( $k$ ) in the investigated flock are  $21.27 \cdot 10^{-3}$  and 13.21, respectively. The value of the Tajima D test (-1.4427,  $p > 0.10$ ) does not indicate a lack of alleles, nor does it indicate a narrowing of the genetic diversity occurring in the history of the population.

Keywords: bimodal mismatch distribution, maternal origin, breed variant

### Introduction

After RODICZKY (1904), the Tsigai (cigája or berke) sheep first came to the historical territory of our country at the end of the 18th century (1792). The demand

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of the domestic post factories and Brassó's (Braşov, Kronstadt) flourishing wool trade encouraged the farmers of the Transylvanian counties to replace their herds of coarse woolly Țurcană (gyimesi racka) with finer (albeit 31-42 micron, "crossbred") wool-producing Tsigai (ESZES and GÁSPÁRDY, 1997). It was SZENTKIRÁLYI (1885) who was the first and corrected to state that the fleece of the Tsigai sheep consists entirely of wool fibres; therefore, it is not a mixed wool sheep, as many people mistakenly claimed.

According to SCHANDL (1941), "sharply defined sub-breeds and ecotypes are not usually distinguished in the Tsigai. Nevertheless, it can be established that the nutritive power of the nugget makes its impact felt". The existence and different origin of the Tsigai became aware of those who dealt with the breed later (VERESS, 1996; DUNKA, 1997; KESZTHELYI, 1997; TÓTH, 1997; KÓSA, 1998).

The colour variants of the Tsigai breed can be separated based on the colour of the fleece and the short hairs. According to the colour of the head and legs, individuals with black, dark brown, light brown, yellowish-red, white and variegated (spotted) heads and legs were distinguished among the white fleecy Tsigais. Spots were more common on the legs than on the head. In the course of history, two basic colour variants of the white-wool Tsigai, also belonging to the mountain type, spread in the Transylvanian parts.

One of them, on SZENTKIRÁLYI's (1923) unifying proposal, is the variant with a completely black head and legs. This spread in other Hungarian historical areas: the Uplands, the South, the Great Plains and, to an extent, the Transdanubia. Several more sub-variants were separated in the Southern Region: the triple-purpose but meatier variant of Čoka (csókai) and the dairy variant (with a milk production of over 100 litres) of Sombor (zombori). Further on, there were the ancient variants, the Tsigai Rumska (árpatarlói cigája; ULMANSKI, 1922) which is now extinct and most reminiscent of Transylvanian Tsigai, and the large-bodied, also with a high yield of milk, the Doroslovo Tsigai (doroszlói cigája; KOVACS, 2000). All of them have chocolate brown or black head and legs, and white fleece. In the territory of today's Hungary, it is worth distinguishing two sub-varieties according to origin: one is the lowland one, which shows the greatest similarity with the variant of Čoka, and the other is the mountain one, which entered Jákotpuszta from the Highlands three decades ago. Almost without exception, they have white fleece and dark brown or black heads and legs.

The other one, is the brown-faced and short-legged, so-called Covasna Tsigai (kovásznai vörösképpű cigája, in Hungarian; ruġine, rusty or red-faced, in Romanian; KUKOVICS, 2006). The first, most classic and almost yellow individuals of this variant were the Tsigais from Hétfalus. Later, HAMMOND et al. (1961) also mention the reddish head but white or variegated wool and the completely black Romanian Tsigais.

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The variants of the Tsigai breed group in other countries have very different names, for example, Karbanat in Bulgaria, Ruda in Albania, and Kivircik in Turkey. These are now not detailed, just like the single purpose dairy-type Tsigais.

In the last decades, the yellow-headed colour variants of the Tsigai native to Covasna and Hargitha counties (BODNÁR et al., 2021) have entered our country. Since 2016, the Hungarian Sheep and Goat Breeding Association has treated them as a standalone breed (thus separated from the black-headed Hungarian native Tsigai) under the name Yellow-headed Berke.

The aim of the study is to characterize the maternal background and maternal genetic diversity of the Red-faced sheep of Covasna. For this, the nucleotide sequence of the control region (CR) of the mitochondrial genome (mtDNA) is used. It is assumed that as an enclosure culture, it can serve with a specific genetic pattern left over from the past. With these results, the genetic identification of the haplotypes (families) of this ecotype is revealed and their successful maintenance is established.

## Material and methods

In 2014, the Institute for Farm Animal Gene Preservation (HGI) brought in a small herd of Red-faced sheep of Covasna from Transylvania with the aim of national conservation of the breed. The sampling was achieved from 34 individuals (20 ewes and 14 rams) 2021 and 2022. Of these, 21 come from the HGI's "ex situ" herd (Gödöllő), which were imported from Transylvania. The other 13 samples were collected in Transylvania from 4 different "in situ" herds.

The blood samples were taken from the jugular vein in EDTA blood collection tubes, then the blood inhibited in coagulation was transferred into Eppendorf tubes and frozen at -20 °C the same day. The blood samples were taken from the selected animals as part of routine veterinary procedures for the detection of infectious diseases (e.g. *Ovine brucellosis*). The further use of these samples obtained during general clinical veterinary procedures for research purposes according to Act XXVIII of 1998 and Government Decree no 40/2013 does not qualify as animal testing, so ethical permission was not necessary.

DNA was extracted and purified using the GenElute Blood Genomic DNA kit (Sigma-Aldrich, St. Louis, MO, USA). A previously described primer pair was used to amplify the segment to be examined (HIENDLEDER et al., 1998). The programmable Thermal Cycler 2720-type PCR equipment (Applied Biosystem, Waltham, MA, USA) was used to amplify the DNA sequence. The PCR products were cleaned with the SIGMA GenElute™ PCR Clean Up Kit (Sigma-Aldrich, St. Louis, MO, USA) following the protocol. For the sequencing reaction, a BigDye® Terminator version 3.1 Cycle Sequencing Kit (ThermoFisher Scientific, Waltham, MA, USA) was used as recommended by the manufacturer. The ABI Prism 3130XL Genetic Analyzer (Applied Biosystems, ThermoFisher Scientific, Waltham, MA,

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USA) was used to analyse and detect the sequence data, according to the manufacturer's instructions. Sequence data were analysed using Sequencing Analysis Software 5.1 (Applied Biosystems, ThermoFisher Scientific, Waltham, MA, USA) and subsequently aligned using Sequencher™ 4.1.2 software (Gene Codes Corp, Ann Arbor, MI, USA).

The sequences were evaluated using the DnaSP 6.0 software (ROZAS et al., 2017). The samples were sorted into haplogroups based on the GenBank reference samples (A-HM236174, B-HM236176, C-HM236178, D-HM236180, E-HM236182 (MEADOWS et al., 2005).

## Results and discussion

The length of the aligned CR sequences is 621 bps. In the entire study sample, the number of monomorphic base sites was 533, while the number of polymorphic base sites was 88 (with singleton variable sites and parsimony informative sites 35 and 53, respectively).

The number of haplotypes is 30. This means that the 34 individuals almost without exception represented their own haplotype. Eight individuals, which are identical shared a pairwise haplotypes (that is, belonging to the same maternal line, not necessarily siblings). Individuals can be classified into two haplogroups. The presence of haplogroups A and B proves the origin of the Red-faced sheep of Covasna in Asia Minor.

The haplotypes of the individuals transported to Hungary match well with the haplotypes of the Transylvanian animals, in some cases falling into the same haplotype. The haplotype distribution of the Hungarian individuals therefore represents the breed well and is a basis for preserving genetic diversity.



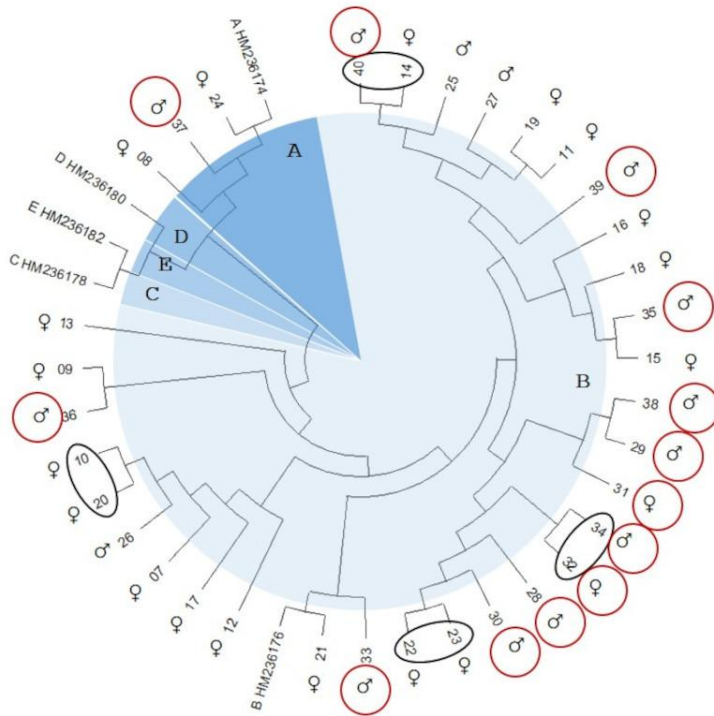


Figure 1. Composition of Red-faced sheep of Covasna according to CR haplotype and haplogroup, with GenBank control haplogroups ((A-HM236174, B-HM236176, C-HM236178, D-HM236180, E-HM236182; MEADOWS et al., 2005). The solid line connects individuals belonging to the same haplotype and are genetically identical. The individuals circled in red live in Transylvania.

The value of haplotype diversity ( $H_d$ ) is 0.991, and its variance is 0.010. The mean of the nucleotide diversity ( $\pi$ ) and the average nucleotide difference per pair ( $k$ ) in the investigated flock are  $21.27 \cdot 10^{-3}$  and 13.21, respectively. Nevertheless, the nucleotide difference is around 10 in most of the herd, and around 35 in the remainder (Figure 2). The dashed line represents the expected distribution for a constant stock size; its course is moderate. Points connected by a solid line reflect a bimodal observed distribution. The distinct peaks indicate that there are two dominant groups of haplotypes associated with a relatively constant population size of the Red-faced sheep of Covasna over time.

The  $D^*$  and  $F^*$  tests of FU and LI performed on the entire test sample did not give significant results, -1.1246 ( $p > 0.10$ ) and -1.4632 ( $p > 0.10$ ), respectively. Unlike these, the value of the FU's  $F_s$  statistic of -13.813 was significant ( $p < 0.001$ ). The value of the Tajima D test (-1.4427,  $p > 0.10$ ) does not indicate a lack of alleles, nor

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does it indicate a narrowing of the genetic diversity occurring in the history of the population.

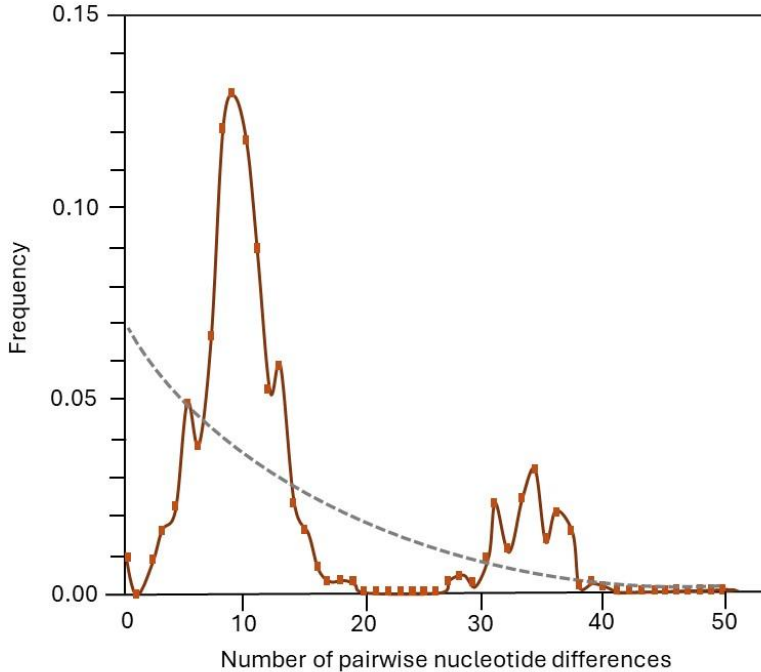


Figure 2. Frequency distribution of the number of sequence mismatches between pairwise combinations of CR haplotypes of Red-faced sheep of Covasna

## Conclusion and recommendation

The first such processing of the Red-faced sheep of Covasna was successfully carried out. The haplogroups A and B determined prove the historically known origin of the Tsigai in Asia Minor, including the Red-faced sheep of Covasna. The breed came to Hungary from Turkey via the Balkans. Haplogroup C has been identified in other Hungarian native sheep breeds. Thus, the breeds like Cíkta (GÁSPÁRDY et al., 2022) and Yellow-faced sheep of Kecskemét (TULLY et al., 2023) have a more complex maternal background.

The number of haplotypes in the studied population is high. This means that almost every individual in the flock belongs to a different maternal lineage, i.e. the maternal genetic background is advantageously diverse. The pedigree of the imported stock is incomplete; therefore, knowledge of the maternal background is essential for

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pairings. Haplotype diversity (Hd) is also high. These observations can also be paralleled by the strongly negative value of Fu's  $F_s$  statistic, which indicates a remarkable excess number of alleles. Together, these facts create the possibility of maintaining the genetic diversity of the breed.

### Acknowledgement

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## Pasture farming as a sustainable method of goat milk and dairy product production

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### Abstract

Pasture represents an economical and sustainable way of feeding goats. However, to fully leverage the benefits of grazing, farmers must manage several aspects, including plant diversity, potentially poisonous plants, pasture load, and the pasture's nutritional value. Moreover, combined grazing with goats, sheep, and cows is the most effective way to utilise pasture resources. Pasture-based diets impact milk and dairy products' nutritional and fatty acid profiles. When goats graze on diverse pastures, their diet typically includes a range of plants, such as grasses, legumes, and herbs, which enhance the nutritional content of their milk. This dietary diversity increases the presence of protein, fat, and beneficial fatty acids, such as omega-3 and conjugated linoleic acid (CLA), both of which are associated with some health benefits. Additionally, pasture-based diets contribute to higher antioxidant levels in goat milk, including vitamins like A and E, which improve the milk's oxidative stability. Grazing improves goat's milk's nutritional and sensory quality, providing a natural method to enhance its value for consumers and dairy processors.

Keywords: pasture management, goat milk, dairy product, added value

### Introduction

Pasture is often regarded as an economical source of feed and nutrients. However, pasture use does not always guarantee adequate production levels, especially due to

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seasonal variations and climatic conditions that affect grass availability and quality (BONANNO et al., 2008). There is no universal grazing system that fits all situations; the effectiveness of different grazing methods depends on achieving optimal interaction between pasture and grazing animals, shaped by specific practical circumstances (BONANNO et al., 2008). Livestock grazing has positive effects on pasture biodiversity, as well as on animal health and welfare (WROBEL et al., 2023). Additionally, grazing significantly impacts the quality of goat milk and dairy products, influencing their nutritional profile and sensory characteristics (PASKAŠ et al., 2023). When goats graze on diverse pastures, their diet includes a variety of plants such as grasses, legumes, and herbs. They can select tannin-rich woody forage and a wide range of plant species, which can enhance forest rangeland biodiversity under rational forage resource management (CHEBLI et al., 2022). Year-round vegetation suitable for goats can be achieved by cultivating a mix of warm- and cool-season grasses (both annuals and perennials), legumes, and forbs, as well as by incorporating browse plants into the grazing system. Browse adds dietary variety, minimizes gastrointestinal parasite issues, and improves goat health and performance. Browse can be integrated into grazing systems by developing woodland grazing plots or by planting browse in designated paddocks (KARKI, 2020). Effective pasture management for goats requires understanding their grazing behaviour, selecting optimal plant species, employing appropriate grazing techniques, and determining the start, end, and frequency of grazing. Grazing frequency depends on plant height, which varies with seasonal grass growth, grazing intensity, pasture load, and the timing, quantity, and quality of supplemental nutrition. Moreover, an efficient grazing system should also protect the environment and landscape (BONANNO et al., 2008).

## Efficiency of pasture utilisation

### 1. Pasture load and poisonous plants

The degree of pasture loading affects its utilization efficiency, but to increase profitability, this loading is often raised irrationally. In a study by ANIMUT et al. (2005), an increase in pasture loading reduced forage mass post-grazing, increased the proportion of grasses, and lowered the nutritional value of the available forage, though these effects varied with climatic conditions. Goats displayed neither a strong preference nor aversion to ragweed (*Ambrosia spp.*), indicating it was neither highly favoured nor rejected compared to other plants. However, both goats and sheep showed a higher preference for ragweed when pasture loading increased. As the loading level rose, the average daily gain slightly decreased, a trend more pronounced in sheep than in goats. Higher pasture loading also raises the risk of animal poisoning. Many plants employ secondary chemicals as defence mechanisms; these are toxic substances, but their effects depend on

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concentration and dose. Toxic compounds in plants include alkaloids in red canary grass and lupins, tannins in clovers and lespedeza, terpenes in fennel and other bitter plants, and endophyte toxins in marsh plants (PROVENZA et al., 2003). Although most plants, including grasses, contain toxins, they generally do not harm animals if consumed in limited quantities. Combined grazing reduces the risk of poisoning from plants like bitterweed (*Actinea odorata*) and sacahuista (toxic in flowers and fruit for cattle, sheep, and goats, though leaves can provide nutrients during sunny periods). In continuous grazing systems with high pasture loading, annual mortality from poisoning was 4% at high load levels, 3.3% at medium, and 1.7% at low. Poisoning was highest among goats grazed alone (5%), decreased with cattle-goat grazing (2.5%), and was lowest in mixed grazing with cattle, sheep, and goats (1.5%). Pastures should be free of poisonous plants like rhododendrons and azalea bushes, which can be fatal to goats. Goats should also be kept off clover- or alfalfa-rich pastures that are wet from rain or dew, as this can cause bloat. While certain plants are poisonous to some animals, they may be safe for sheep and goats. For instance, milkweed (*Euphorbia spp.*) causes hoof skin loss, digestive irritation, and sometimes death in cattle, but provides 20-27% protein for sheep and goats in season. The yellow star thistle, which is toxic to horses and can cause "chewing disease," has no negative effect on cattle, sheep, or goats (PASKAŠ, 2022). Additionally, poisoning in goats can result from plants containing grayanotoxins (*Pieris and Rhododendron species*) or cyanogenic glycosides (e.g., cherry laurel, *Prunus laurocerasus*). Other plants known to cause goat poisoning include yew (*Taxus species*), and oak bark (*Quercus spp.*), hemlock (*Conium maculatum*), leylandii (*Hesperotropis leylandii*), box (*Buxus sempervirens*), and those with cardiac glycosides, such as oleander (*Nerium oleander*), or pyrrolizidine alkaloids, such as ragwort (*Jacobaea vulgaris*) (BATES, 2022).

### **1. The botanical structure and usage of a pasture**

Sustainable goat production on pastures involves optimizing pasture use while reducing dependence on cereals and cultivated forage (PASKAŠ, 2022). Feed composition and digestibility impact dry matter intake, with high structural carbohydrate content and low organic matter digestibility generally reducing intake in both confined and pasture-fed animals. However, in grazing small ruminants, the relationship between intake and plant chemical composition may involve more complex mechanisms (PULINA et al., 2013). Animals have an innate sense of which plants are beneficial for them to consume. When grazing freely, animals quickly select the more palatable plants, allowing fewer desirable species to spread rapidly. Upon arriving at a new area, goats graze actively, but over time, they slow down, leaving large portions unused, which can lead to an increase in weeds (MEMIŠI et al., 2011). Excessive grazing, on the other hand, can cause erosion; therefore, controlled and planned grazing is recommended to prevent both underuse and overuse of pastureland. Neglecting pastures allows undesirable species,

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including aggressive neophytes like *Amorpha fruticosa*, to proliferate. While young *Amorpha* plants are susceptible to trampling and can be grazed, they become woody and unusable as they mature, altering the native flora. Other invasive shrubs, such as juniper (*Juniperus sp.*), blueberry (*Vaccinium sp.*), acacia, and hawthorn, also grow quickly and more vigorously than native plants, turning pastures into dense, unproductive thickets. Certain plants, like Chinese lespedeza (*Sericea lespedeza*) and bird's-foot trefoil (*Lotus corniculatus*), contain high levels of condensed tannins. *Sericea lespedeza* thrives on poor, acidic soils and helps control internal parasites, reducing parasite load in sheep and goats. Goats and sheep may require time to acclimate to this plant, while cattle will consume it if it is not overly mature. Other plants with similar effects include chicory, grape seeds, cranberries, pine bark, and others (PASKAŠ, 2022).

The protein level in the diets of pastured goats depends on the botanical composition of the pasture and the ratio of shrubs to grassland. While shrubs and certain plants provide nutritional benefits, they sometimes contain secondary metabolites—such as tannins, phenols, and terpenes—that can repel animals or be toxic. Tannins, a complex of high-molecular-weight polyphenolic compounds, are better tolerated by goats. Goats have mechanisms to neutralize the antinutritional effects of tannins, including salivary secretion with high proline levels and ruminal processes. Additionally, ruminal fermentation and the adaptation of ruminal microflora to terpenes enable goats to more effectively utilize terpene-rich feed (LANDAU et al., 2000). Generally, when additional energy and protein sources are available, goats can consume more plants containing tannins, terpenes, and saponins (PROVENZA et al., 2003). Although selected shrubs in research by CHEBLI et al. (2022) contained a high level of condensed tannins, goats consume them readily. By selectively grazing, goats generally consume plants richer in digestible organic matter, crude protein, and sugars while avoiding those higher in neutral detergent fibre (NDF) (PULINA et al., 2013). Proper pasture management enriches the soil by adding organic matter. Since goats typically consume grasses (e.g., ryegrass, fescue, and sea buckthorn) and some broadleaf "weeds" (e.g., chicory, wild carrot, and sweet potato), with less emphasis on legumes (e.g., clover and alfalfa), pasture improvement efforts should focus on increasing vegetation density and enhancing soil fertility. During summer, goats can meet part of their nutritional needs by consuming plant species commonly rejected by other ruminants, such as *Rumex* spp., *Daucus carota*, and *Convolvulus arvensis* (BONANNO et al., 2008). Goats can graze plants at heights between 20 and 120 cm. High-quality cultivated pastures for goats include clover, with a recommended grass-to-legume ratio of 1:1. The optimal plant height on pastures is 8–12 cm; when available pasture mass falls below 1,000 kg/ha, goats tend to stop grazing (GRBEŠA et al., 2005). In a study by JORGENSEN et al. (2007) comparing goat grazing on natural and cultivated pastures (with *Phleum pratense* [Timothy] and *Festuca pratensis* [meadow fescue]), goats showed a preference for Timothy early in the summer (85% of their diet) but



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transitioned to a more varied diet later in the season, combining Timothy and fescue with couch grass (*Elytrigia repens*) and tufted hair grass (*Deschampsia cespitosa*). On natural pastures, goats had a more diverse diet, with access to shrubs, leaves, grasses, and ferns, among others. At the end of summer, with a decline in protein content, they spent more time in vegetated pasture areas while avoiding boggy, thorny patches. Benefits are more substantial in mixed grazing systems where goats graze with other ruminants, especially when animals have higher nutritional requirements and exhibit diverse feeding behaviours. Differences in selective feeding between sheep, cattle, and goats make these systems more efficient, significantly influencing the botanical and structural composition of pastures (BONANNO et al., 2008). Studies also showed that combining cattle, sheep, and goats on a single pasture can increase pasture utilization by about 25% (COFFEY, 2001).

## **2. Pasture quality and supplements**

Pasture quality is influenced by various factors, with plant composition and nutrient content significantly impacted by the physiological stage of plant maturity (PASKAŠ, 2022). TUDISCO et al. (2010) observed that pasture quality was poorest in July, while in May and September, protein content was higher and NDF levels were lower. Similarly, in research by PASKAŠ et al. (2019) was observed that in May and June, pastures contain the most soluble nutrients, inversely correlated with crude fibre content. CHEBLI et al. (2022) also found significant seasonal variations in forage availability, quality, and feeding behaviour. However, they noted that seasonal changes did not significantly affect the forage preferences of indigenous goats. LENG (1990) categorizes pastures as low to medium nutritional quality if they contain less than 55% organic matter and 8% crude protein. During summer, fermentable compounds (especially sugars) decrease significantly, while lignin content rises, further reducing the pasture's nutritional value (BONANNO et al., 2008). Legumes and grasses show contrasting values in digestibility and intake. Under similar growth conditions, legumes contain less NDF but have highly lignified cell walls, whereas grasses exhibit high NDF content and lower lignification. This results in comparable digestibility for grasses with higher NDF but lower intake for equal digestibility levels (VAN SOEST, 1965). Young pastures with easily digestible fibre offer higher energy levels (GRBEŠA et al., 2005). According to MORAND-FEHR et al. (2007), pastures alone may not always meet all of a goat's nutritional requirements, necessitating supplemental concentrates. However, accurately assessing actual pasture intake is challenging, making it difficult to determine the required concentration amounts. Seasonal variations and climate differences further complicate assessing the quality and availability of pasture biomass and predicting when supplemental concentrates might be needed. For goats grazing on both natural and cultivated pastures, concentrates generally help increase energy intake and improve milk production

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(BONANNO et al., 2008). Cereals rich in starch and energy, such as corn, barley, oats, and wheat, form the core of concentrated supplements. Goats, however, can struggle to adapt to high-concentrate diets, with rapid changes increasing the risk of acidosis, enterotoxemia, and urinary calculi. While young, leafy pastures can often meet goats' protein requirements, ruminants also need energy to utilize nitrogen from plant proteins for microbial protein synthesis, which requires additional energy from concentrated nutrients (GRBEŠA et al., 2007). Energy deficiency in grazing goats can result from their physical activity. For this reason, and to improve productivity, the addition of concentrated feed and mineral-vitamin supplements is generally recommended. However, concentrate supplementation can reduce the time goats spend grazing. LANDAU et al. (2000) observed that while high levels of concentrate aid body condition recovery in grazing goats, they also decrease grazing activity. Goats receiving small amounts of supplements spent more time grazing than those given larger amounts (75% vs. 59% of total observation time, respectively). FEDELE et al. (1993) observed that pasture intake decreases as concentrate supplementation increases, particularly when supplements are high in crude protein. Goats receiving 150g of concentrate with 18% crude protein consumed 51% more forage than those given 550g daily, suggesting self-regulation based on protein needs. Additionally, each kilogram of concentrate can reduce roughage intake by 0.5–0.6 kg of dry matter (GRBEŠA et al., 2005). While high-quality pasture generally meets protein requirements, concentrate supplementation is beneficial when pasture quality is low, as it supports milk yield. To optimize feeding, a maximum of 600g of concentrate per lactating goat per day is recommended (BONANNO et al., 2008).

### **Influence of pasture feeding on milk quality**

Numerous studies have confirmed the positive influence of pasture feeding on the fatty acid composition of goat milk (YAKAN et al., 2019; OTARU et al., 2020; PASKAŠ et al., 2023). Natural pastures provide significant benefits for goat dairy products compared to confinement systems, especially in the early stages. GIORGIO et al. (2019) noted that grazing on diverse grasses and legumes enhances the dairy profile with beneficial polyphenols and fatty acids. Furthermore, grazing livestock on diverse pastures concentrates a greater variety and higher amounts of phytochemicals in meat and milk compared to grazing on monoculture pastures, while phytochemicals are further reduced or absent in meat and milk from grain-fed animals (VLIET et al., 2021). Pasture-raised goats produce milk with higher levels of polyunsaturated n-3 linolenic acid (3.32% compared to 1.21%) and a more favourable fatty acid profile, including a lower n-6/n-3 ratio. In addition, healthier indices were observed, including lower atherogenic (AI) and thrombogenic indices (TI), as well as higher health-promoting index (HPI) and hypo-

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/hypercholesterolemic(HH) values. In addition, a higher cheese yield of 22.24% was observed compared to confinement (PASKAŠ et al., 2023). OTARU et al. (2020) also confirmed that higher levels of concentrates increase short and medium-chain fatty acids in cheese, while lower levels of concentrates increase long-chain, polyunsaturated, ruminal and n-3 fatty acids, resulting in a lower n-6/n-3 ratio. Research shows that milk from pasture-raised goats has lower saturated fatty acids and a more favourable fatty acid profile, especially in early lactation (YAKAN et al., 2019; CURRÒ et al., 2019). Pasture feeding also influences the quality of cheese and other dairy products. Thus, cheese from pasture-fed goats had lower fat and cholesterol content (12.3g/100g and 63.2mg/100g vs. 16.9g/100g and 80.4mg/100g) and higher tocopherol content (211mg/100g vs. 87mg/100g), with a favourable n-6/n-3 ratio and improved sensory qualities such as colour, texture, aroma and characteristic "goat smell" (GALINA et al., 2007; BARLOWSKA et al., 2018). It also had higher dry matter and protein content, with lower values of AI and TI indices, and higher HPI and HH (BARLOWSKA et al., 2018; PASKAŠ et al., 2023). In addition, grazing resulted in higher concentrations of vitamin A (0.026 vs. 0.036 mg/100 ml) and D<sub>3</sub> (0.075 vs. 0.089 mg/100 ml) compared to hay feeding (PAJOR et al., 2014), and several studies have found increased antioxidant activity in meat and milk of grass-fed vs. grain-fed animals (VLIET et al., 2021).

## **Conclusion and recommendation**

Pasture quality is influenced by various factors, including plant composition and nutrient content. The botanical composition of natural pastures may fluctuate throughout the year, potentially failing to meet all of a goat's nutritional needs. Supplementary concentrates are therefore often required, although their use may reduce the amount of time goats spend grazing. Nevertheless, pasture feeding remains one of the most economical and natural ways to sustain goat production, contributing to the sustainable development of this industry. This practice positively affects not only the nutritional value of goat milk and dairy products but also enhances sensory characteristics, with biochemical pathways contributing to unique flavours. Studies show that grazing animals tend to produce milk with slightly higher fat and protein content, which improves its suitability for cheese production. However, these benefits depend on factors such as pasture quality, availability, and seasonal variations.

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## **Biotechnology assisted approaches in the preservation of rare breeds' diversity**

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### **Abstract**

Preservation is defined within the concept of conservation. It is an aspect of conservation, by which a sample of an animal genetic resource population is designated to an isolated process of maintenance, by providing an environment free of human forces, which might bring about genetic change. The process may be run *in situ* (*in vivo*) in natural environment; it may be run *ex situ* (*in vivo*) or cryogenic storage (*in vitro*). The *in libro* concept of preservation is the conscious “keeping alive” of the one-time presence of an already extinct domestic animal breed or all the characteristics.

When the term "biotechnology" was coined, it meant the process, by which raw materials can be biologically transformed into socially useful products with the help of living organisms (fermentation). The production of special foods that are matured in a controlled manner from the milk and meat of old domestic animals may be an area to be exploited more strongly in the future. Later, reproductive biotechnology, several biotechnical procedures have been developed, such as embryo transfer (ET) and artificial insemination (AI). There are many other reproductive techniques, ranging from cloning to sperm and embryo cryopreservation, including hormonal control of reproduction (e.g., induction of cyclicity and parturition). These biotechnological methods are already widespread in everyday animal production and have gained a reasonable right to exist in breed maintenance as well.

The applications of biotechnology are diverse, and it is necessary to use the areas of its application that result in the legal and effective preservation of genetic diversity of rare animals.

Keywords: genetic diversity, food fermentation, reproductive techniques, genetic engineering of animals

## Introduction

The manuscript is a written version of the plenary presentation on the 8th Winter School on Animal Biotechnology 2024 with topic „Animal Biotechnology and Biodiversity” (in AgroBioTech Research Centre, Slovak University of Agriculture in Nitra, Slovakia), 1st February 2024.

Considering the title of the event, I chose the topic of my presentation. By connecting the two activities, we can review the options and procedures that can be used to bring the preservation of our old, endangered domestic animal breeds up to date.

At the beginning, I would like to briefly define the concepts of conservation and preservation.

The definition of conservation (of genetic resources) can be given as follows: The management of human use of the biosphere so that it may yield the greatest sustainable benefit to present generations while maintaining its potential in concordance with the Convention on Biological Diversity of Rio de Janeiro, 1992. It tells about the fair and equitable sharing of the benefits arising from commercial and other utilization of genetic resources. This activity includes preservation.

The preservation aspect of maintenance by which a sample of an animal genetic resource population is designated to an isolated process of maintenance, by providing an environment free of human forces which might bring about genetic change.

This may even include wild species threatened by extinction, but we are now narrowing down this definition to endangered domestic animal breeds.

At the same time, we immediately see its three versions (*in situ*, *ex situ in vivo*, and *ex situ in vitro*), in which biotechnology can play a role.

When a separate section was created in the EAAP for breed preservation first (in the 90s), the young people gave the definition of the term: a session of EAAP where old people talk. Since then, the situation has improved a lot, thanks in part to the rise of biotechnology.

When the term "biotechnology" was coined, it meant the process by which raw materials can be biologically transformed into socially useful products with the help of living organisms (fermentation).

Later, new branches of this were formed, which even lack the intermediate (micro)organism. In some of these, sub-organismal units of a living being are used, in others, it is purely about technical operations and organizational issues.



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In 1989 Robert Bud gave account of the fact that the father of the term "biotechnology" was the Hungarian agricultural engineer, Karl Ereky (1878-1952), 1919.

No such "father" has been named in the field of breed maintenance. In any case, one of its prominent representatives was Imre Bodó (1932-2023), who died in December. I would also like to project his personality, because today's generation can learn a lot from his work.

According to another funny definition, which became winged words in Hungary biotechnology is "what László Kállai (1927-2007) gives money for". He was the curator of the distribution of application resources in the 1980s, when more and more biotechnology applications were submitted from the field of agricultural science.

Biotechnology has numerous applications, particularly in medicine and agriculture. So, we can group this according to the nature of the utilization area.

This can be arranged according to the objectives to be completed (e.g., healing and production).

A reasonable resolution can be made according to the means (methodology) of the process. For example, exploring the use of microscopic equipment that can enter the human body (nanotechnology), next to embryo splitting.

In addition, the direct biotechnology processes are surrounded by ancillary scientific fields and miscellaneous supplementary tasks for the sake of completeness. Like bioinformatics. Safety precautions fall under two principal headings, occupational safety and public safety.

Finally, we must mention the consequences of biotechnology procedures. We do not yet know much about their direct and indirect forms. We have significantly different opinions on the application of biotechnology procedures. Whereas U.S. regulation of GM foods is based on the product, European Union (EU) regulations are based on the process. As a result, the EU regulates GM plants and animals more stringently, and European publics are wary of genetically engineered foods.

Today, eight branches of biotechnology are distinguished, which are also usually displayed in different colours.

Red biotechnology involves medical processes, such as using organisms to produce new drugs and stem cells to regenerate damaged human tissues and grow and regrow entire organs.

White or grey refers to industrial processes, such as the development of new chemicals or new biofuels for vehicles. Additionally, the environmental biotechnology which application is to develop sustainable environmental practices that reduce pollution and waste. (The following are its examples: phytoremediation uses genetically engineered microorganisms to purify soils of heavy metals and other pollutants. Bioremediation. Plastic-eating bacteria breaks down waste such as plastic in soils and water.

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Blue encompasses processes in marine and aquatic environments, such as converting aquatic biomass into fuels and pharmaceuticals.

The following two branches of biotechnology are related and affect us the most due to our profession.

Green covers agricultural processes, such as producing pest-resistant crops, disease-resistant animals and environmentally friendly agricultural practices.

Yellow refers to processes that aid food production. Approximately 6,000 years ago, humans began to tap the biological processes of microorganisms in order to make bread, alcoholic beverages, and cheese and to preserve dairy products.

Gold, also known as bioinformatics, is a cross between biological processes and informatics. It refers to the methods researchers (healthcare workers) use to gather, store and analyse biological data to solve problems (treat patients) and make useful products. Here we can cite the precision farming and the use of artificial intelligence. Most notably, the singularity would involve computer programs becoming so advanced that artificial intelligence transcends human intelligence, potentially erasing the boundary between humanity and computers.

Legislation in violet ensures the practice of biotechnology is in compliance with laws and ethical standards governing each field.

Biotechnology also comes with misuse and disadvantages. The former one is shown in dark or black which is the use of biotechnology for weapons or warfare.

The next slide, partly as a continuation of the previous one, presents the main disadvantages of biotechnology, which are as follows: biological warfare, high costs, safety questions, ethical considerations.

Concerns about biotechnology's disadvantages have led to efforts to enact legislation restricting or banning certain processes or programs, such as human cloning, GMOs and embryonic stem-cell research.

The disadvantages include the reduction of soil fertility and biodiversity.

Seeing this, one can question whether we can really use biotechnology to preserve genetic diversity and old domestic animal breeds.

If the answer is “yes”, it is obvious that the application area of biotechnology in the preservation of old breeds is the agriculture, including animal husbandry.

When the term "biotechnology" was coined, it meant the fermentation. The production of special foods that are matured in a controlled manner from the milk and meat of old domestic animals may be an area to be exploited more strongly in the future.

Biotechnology gradually gained appreciation in the reproduction of our old rare breeds as well.

Several biotechnical procedures have been developed for both the female and male side. Such as embryo production (ET) and artificial insemination (AI). There are many other reproductive techniques, ranging from cloning to sperm and embryo cryopreservation, including hormonal control of reproduction (e.g., induction of cyclicity and parturition). These are the biotechnological methods that are already widespread in everyday animal production and have gained a reasonable right to exist in breed maintenance as well.

In-depth characterization of a breed now undoubtedly includes direct DNA testing by use of microsatellites, SNPs, longer and shorter DNA segments- and whole genome sequencing. Moreover, in some cases, QTL association analyses and gene expression studies. Soon, genomic breeding value will also be estimated in the old breeds. The molecular genetic screening of monogenic defects is also spreading in these breeds. It is important to check pedigree data, paternal lines and maternal families with molecular genetic tools. It is urgent to introduce molecular genetic control of parentage and individual identity and food chain, to calculate coefficients of molecular inbreeding and degree of kinship.

Well, now let's take a look at the preservation variants which can now be considered crystallized. Certain biotechnological methods or biotechnical procedures have already been used, or may be used in the future in these. The variant may be *in situ* whereby the pool consists of live animals (*in vivo in situ*) in natural environment. During the maintenance of endangered animals *in situ*, we must pay attention to the regular, original use (in traction, and raw material and food production), breeding, feeding, and housing of the animals.

It may be *ex situ*, whereby the sample is placed, for example in rescue station (*in vivo ex situ*, this is called as the “necessary but wrong solution” in quotation marks, but not at all (test station, centralized sire rearing, zoos etc.) or cryogenic storage (*in vitro ex situ*). The *in libro* concept of preservation is the conscious “keeping alive” of the one-time presence of an already extinct domestic animal breed or characteristics lost by today and remaining knowledge of a still living rare breed in our common knowledge.

Looking back from today's time, it is very interesting that the preservation of *tissues and genes* separately was envisioned decades ago among *in vitro* methods.

I note that the concept of breed preservation is often replaced by gene conservation in public life. In any case, if we indicated the preservation of separated genes in this form, then this already gained the right to exist. Genes are transferred from one breed to another with traditional crossing *in vivo* (*gene introgression*) and preserved in the recipient breed with accelerated back-crossing (MAS – *marker assisted selection*). An example is the creation of genetically hornless Limousine cattle in the practical breeding or congenic strains in the laboratory animal science.

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The *gene rescue*, an additional, novel element of domestic breed maintenance program, is based on this principle. Which is strongly connected with the *in libro* version. In other words, if we find a trait believed to have disappeared in an individual of a country herd, we can return it to the breed in question, from which this trait has disappeared (example: winter hair coat whitening of horse).

Another important technique used in biotechnology includes *tissue culture*, which allows us to grow cells and tissues in a laboratory for research and medical purposes. From the point of view of breed maintenance, it can be an interesting, sub-organismal unit for the storage of genetic resources (gene conservation). If necessary, these (embryonic) cells merged (blastocyst fusion) can develop into an organism (a breed identical chimera). *Mitochondrial replacement therapy* (MRT) and *three-parent in vitro fertilization* (IVF) introduce more genetic information. It draws attention to the special treatment of multiparental inheritance (crossed maternal lines, *heteroplasmy*) and *epigenetic effects*.

Most novel, and at the same time in the animal production most critical key technique of biotechnology is *genetic engineering* allowing us to alter the genetic makeup of organisms to realize the requested properties.

At the very theoretical level of breed maintenance, it is possible to consider the precise introduction of the desired allele of a genetically healthy breed mate into another one which requires modification of its dysfunctional or harmful allele (breed identical transgenic animal).

Melioration may include insertion/replacement of genes responsible for resistance (heat, parasites etc.).

Possibly applying techniques of stem cell research and cloning to replace dead or defective cells and tissues (regenerative medicine).

Genetically modified organism (GMO) foods of plant origin (e.g. tomato) stay fresher longer and reduce food waste.

## **Conclusion and recommendation**

Today's opportunities and needs result in a certain amount of change. The applications of biotechnology are diverse, and it is necessary to use the areas of its application that result in the legal and effective preservation of genetic diversity of rare animals.

References are available at the Author.

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## **The endangered indigenous breed – the Drežnica goat as a symbol of local identity in Slovenia**

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### **Abstract**

Historically, goat farming was an important economic activity and a significant source of income, especially for the inhabitants of the western part of Slovenia in the Julian Alps (Posočje<sup>†</sup>). In this region, dairy cattle were predominant at middle altitudes, while goats were only kept in the higher mountain areas, where the steep and rocky mountain pastures were only suitable for goats. Over the centuries, this environment gave rise to the indigenous Slovenian goat breed, the Drežnica goat. Even today, the Drežnica goat is closely associated with traditional agricultural practices such as seasonal dairy farming, while the chevon type of the Drežnica goat is characterised by an even more specific type of agricultural practise that is unique to Slovenia. In Slovenia, goat farming and grazing were often accompanied by legal restrictions in the past, which led to the depopulation of mountain villages and the transformation of the landscape. This study underscores the resilience of the Drežnica goat breed, highlighting its crucial role not only in sustaining biodiversity but also in maintaining the cultural and ecological heritage of the Posočje region.

Keywords: Drežnica goat, restriction of goat breeding, intangible cultural heritage

### **Introduction**

A few decades ago, the environment was affected by globalisation, urbanisation, population growth, global warming, and climate change. The loss of indigenous

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<sup>†</sup> Posočje is a geographical area in western part of Slovenia, which includes the territory along the Soča river.

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(native, autochthonous, or local) breeds and animal genetic resources (AnGR) in general is a major problem for food and nutrition security. In recent decades, the number of indigenous breeds has declined due to the demands of intensive livestock farming and global economic development. It is estimated that almost 30% of indigenous breeds worldwide are endangered (FAO, 2021).

In Slovenia, the conservation of AnGR is included in the various sectoral strategies, plans, and programmes at national level. The long-term programme for the conservation of AnGR biodiversity serves as a strategic document containing the priority measures needed to protect AnGR, with a focus on indigenous Slovenian breeds (Programme, 2023). National legislation recognises 14 livestock breeds as indigenous to Slovenia, but only one of them is the indigenous Slovenian goat breed – the Drežnica goat. This breed is highly valued for its excellent adaptability to poor conditions (Figure 1).



Figure 1: Drežnica goat

The Drežnica goat has the smallest population size among the indigenous breeds in Slovenia. It consists of 1100 goats and is listed as a highly endangered breed (Register, 2023). The population of Drežnica goats is divided into two types depending on the purpose of breeding in their original habitats. The milk type of the Drežnica goat developed in the Bovec area, where the tradition of cheese production dates back to the 13th century, while the meat type of the Drežnica goat developed in the Drežnica region. Goat herds for meat production are predominant (ŽAN, 2019).

In the past, goat breeding was of great importance on Slovenian territory. This influence can also be seen in the fact that most of the local names of all livestock species are derived from the name goat, and many of them have been preserved: Kozjek, Kozarica, Kozarski potok (the Slovenian name for goat is “koza”) (NOVAK, 1970). However, goat farming was not a respected activity in the past,

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and the derogatory term “poor peasant cow” was often used for goats (ŽAN, 2016). The neglect of goat keeping as a livestock activity resulted from a series of grazing bans and sometimes even bans on keeping goats indoors (ŠALEHAR et al., 2014). Despite the great usefulness of the goat as a farm animal, providing excellent milk, meat, skin, etc., goat farming was neglected as a livestock sector. In forestry, it was felt that goat farming should be restricted as much as possible or even eradicated, as they cause damage to young trees. Thus, over the centuries, goat farming was accompanied by numerous legal restrictions, although some foresters themselves admitted that it was not the goats that were the problem, but the unsystematic logging, which often went as far as "clear cutting". The forests in the area of Posočje were overexploited as early as the 16th century, when large quantities of felled timber were used to supply Gorica and trade with Venice (ŽAN et al., 2023). Ironworks were a major consumer of wood. Fires and later the First World War also significantly impacted forest destruction, destroying large areas in Posočje, and the Second World War had similar consequences. During the occupation, the Italians ruthlessly exploited and plundered our forests, leaving their remains behind. Legal regulations that restricted or even prohibited the breeding of goats are known from various eras. Breeding in stables was also often prohibited. In this way, the poor sections of the population were deprived of the only possibility of surviving in this demanding environment, which led to emigration and the gradual depopulation of the mountain villages (ŽAN et al., 2023). With this article, we want to draw attention to the ethnological, socio-economic, and sustainable-ecological aspects of Drežnica goat breeding, which has a completely different meaning today than in the past.

### **In the fifties of the 20th century, goat breeding was labelled a cultural disgrace**

After the end of the Second World War, the majority of the population in Posočje worked in agriculture (ŽAN *et al.*, 2023) (Figure 2). In 1946, a special regulation was issued for goat breeding, after which the number of goats fell drastically and goats were almost completely eradicated. Further restrictions on goat breeding followed in the following years. Official documents stated that goat farming was a cultural disgrace on the landscape. With the departure of the older generation, who attached body and soul to the farming way of life and work, the composition of households changed more and more in favour of industrial employment. In addition to the goats, the number of other farm animals also declined. The mountains began to become overgrown, and some of them are now completely abandoned (ŽAN et al., 2023).



Figure 2: Goats in Posočje, 1950

### **Goat farming as a survival strategy and the Drežnica goat as a symbol of local identity**

In the past, goat breeding was most widespread in the Slovenian region of Posočje, where, due to the natural conditions, it was an important and often the only source of livelihood for the inhabitants. Posočje is the cradle of Slovenian goat breeding and represents the original territory of the only surviving indigenous Slovenian goat breed, the Drežnica goat.

The restriction of goat farming in this demanding environment has deprived the poor population of their only source of livelihood, which has led to the migration and gradual depopulation of mountain villages and environmental changes in the surrounding area. Posočje is the cradle of Slovenian goat breeding and represents the original territory of the only surviving indigenous Slovenian goat breed, the Drežnica goat. As a specific economic sector of great importance, goat breeding is still present in everyday life today, even though there are fewer and fewer breeders in Posočje (ŽAN et al., 2023) (Figure 3).





Figure 3: Milking the Dreznica goats on the mountain

Breeders born in the first half of the 20th century told us with tears in their eyes about the forced abandonment of goat farming, which led to severe existential and psychological problems (ŽAN et al., 2023). As people were forced to give up farming and work in factories, the future was uncertain and unemployment prevailed. People from the Soča Valley emigrated abroad, mainly to America. Emigration only came to a halt around 1960.

In these places, people lived in balance with nature because, together with the goat, they were completely dependent on its conditions. It was a centuries-old transmission of knowledge about goat farming and the values associated with it, which were greatly appreciated by the people. The consequences of the ban were empty villages and a demographic structure with an ageing population. The consequences of the exodus of people from the villages of Posočje continued decades later and are still visible today. The decades-long suppression of goat breeding after the Second World War is reflected in the selection work of the Drežnica goat, which was, therefore, only established in 2000.

Goat breeding is a specific sector of great importance that is still present in everyday life today, although the number of animals in Posočje is not decreasing as fast as the number of breeders. Thus, breeding is spreading among hobby breeders, for whom goat breeding is not a primary source of income. Goat breeding is therefore not only an agricultural industry, but also an important element in the creation of local identity. The threat to the Drežnica goat jeopardises national biodiversity and intangible cultural heritage.

Apart from the small population of the Drežnica goat, the breed's existence is threatened by the lack of active, professional young breeders and the recent increase in attacks by brown bears, wolves, and jackals. Even the appearance of the disease in such a small area can lead to the extinction of the breed. Together with the Drežnica goat, whose existence is still highly endangered, the national biodiversity

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is also threatened, and with it the intangible cultural heritage with its network of complex knowledge, skills, and values that are passed down from generation to generation in the families and herding communities of the region (ŽAN et al., 2023). Today, people no longer depend on goat farming to the same extent and goat farming is increasingly recognised as a valued local activity. Goat products are sought after by consumers and are often an integral part of first-class culinary offerings. Posočje is home to an internationally recognised Slovenian restaurant with a local menu (including dairy products from Drežnica goats) which has been awarded two Michelin stars.

## Conclusion and recommendation

The harsh measures to persecute goats, especially after the Second World War, had irreversible consequences for the western part of Slovenia called Posočje: depopulation of villages, the psychological harm to residents, the overgrowth of mountains, an altered landscape, reduced biodiversity. Fortunately, rare and courageous breeders with strong attachments to their goats managed to preserve the only autochthonous goat breed in Slovenia – the Drežnica goat, which today represents the local identity of the Posočje region, where over the centuries it has developed into a unique breed fully adapted to the conditions of its environment. In our research, we sought to identify "who" advocated for, enacted, or demanded these stringent measures and laws. Unfortunately, the original documents can no longer be found and it is very likely that they were destroyed after the Second World War. The traditional agricultural practices for breeding the Drežnica goat breed represent an invaluable intangible cultural heritage. Amidst increasing globalization, preserving this heritage is crucial as it highlights Slovenia's cultural diversity. On the other hand, this heritage is the source of a wealth of knowledge and skills that have been passed on from one generation to the next for centuries and are currently threatened with extinction. The social and economic value of documenting and, above all, directly passing on this type of knowledge is important both for goat farming and for the Slovenian state and, last but not least, contributes to the treasury of the cultural heritage of all mankind.

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## Slovak - Hungarian cooperation to protect biodiversity

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### Abstract

The Slovak National Agricultural and Food Centre (NPPC) in cooperation with the National Centre for Biodiversity and Gene Conservation (NBGK) has been awarded a grant under the Interreg VI-A Slovakia-Hungary Cooperation Program for the implementation of its project "Support for cross-border biodiversity conservation in *ex situ* conditions" (HUSK/2302/1.2/018, #Biodiverzity). Its total budget is 998 683,56 EUR, of which 798946,84 EUR is Community funding for the period from 1 May 2024 to 31 October 2026. In the Danube region, due to similar environmental factors, the established indigenous species share common characteristics. Both countries are facing the loss of native varieties and a significant decline in their populations as intensive farming systems become more widespread. Without conscious genetic conservation programs, these varieties could disappear permanently. Therefore, it is a priority for both nations to preserve and integrate the genetic material of these varieties into production. This call for proposals aims to establish the foundations for long-term cooperation between the two countries. By pooling expertise from different fields, sharing knowledge, best practices and innovative solutions, we can jointly achieve our objectives. The project will be implemented collaboratively, led by the Slovak partner. Both countries will contribute resources - financial, infrastructural, and human - to ensure the successful implementation of the tasks.

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## Introduction of the project

The goal of the project is to foster cross-border cooperation and support in protecting biodiversity, with a focus on preserving selected animal genetic resources under *ex situ in vivo* and *in vitro* conditions. This includes the mapping, selection and genotyping of original Slovak and Hungarian animal breeds, which are raised only by small-scale breeders. Due to this, the number of individuals is very low and some breeds are at risk of extinction.

*Ex situ in vitro* experiments will focus on sampling for the following purposes:

- DNA isolation and determining the origin of individuals,
- Collection of ejaculates and blood for microbiological, morphological and andrological examinations,
- Isolation of stem cells from poultry and rabbits,
- Cryopreservation of biological material (sperm, stem cells, bone marrow) from national animal breeds, including the brown hare, for preservation in gene banks.

*Ex situ in vivo* efforts will involve collecting animals from each breed, including 13 rabbit breeds, 1 brown hare breed, 2 poultry breeds and 4 sheep breeds with at least two individuals from each breed.

The project will result in cryopreserved samples stored in gene banks on both the Slovak and Hungarian sides, mutual exchange of reserve samples and a collection of national breed animals. The effective transfer of knowledge to target groups will be ensured through professional events, conferences and publications raised throughout the project.

## Objectives of the NBGK

The Hungarian Giant Rabbit is a critically endangered species. To maintain its genetic diversity, it is essential to increase the number of individuals in the nucleus population at the Institute by introducing new breeding stock based on genetic testing of external breeders. There are limited data available on the breed's spermatological parameters and its semen's tolerance to freezing. In addition to *in vivo* gene conservation, developing an *in vitro* method for deep freezing of semen is crucial.

Among our native poultry breeds, two are critically endangered, ten are under protected status and two are under conservation status. This further justifies the need to develop *in vitro* gene conservation methods for these breeds. While various semen storage protocols exist, our previous studies have shown that the efficiency of semen preservation in our indigenous poultry species often falls below international standards. Therefore, further research is required to identify the causes and improve current preservation methods.

In poultry, female gametes are heterogametic, meaning that the *W* chromosome and mitochondria are located in the oocyte, which cannot be preserved solely through

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sperm freezing. Due to the biophysical characteristics of eggs, they are not cryopreservable; however, embryonic stem cells or day-old gonadal tissue may be suitable for long-term preservation of the female genome. Among the different types of stem cells, primordial germ cells (PGCs) are precursors to sperm or oocytes, can be maintained in cell culture and are cryopreservable, making them ideal for genetic preservation.

The aim of the project is to establish stable male and female cell lines from the Slovak indigenous Oravka breed, which, after *in vivo* and *in vitro* testing, can be stored in the gene bank. Deep freezing of gonadal tissues has already been implemented for several breeds, and the goal is to expand the gene bank samples at our institute while transferring and introducing this method to the Slovak gene bank. Both PGCs and gonadal tissue are capable of recovering 100% of the poultry genotype, preserved in the F1 generation through grafting and artificial insemination post-maturation. The results of this project will contribute to the conservation of genetic diversity in indigenous livestock breeds in both countries.

### **Objectives of the NPPC**

The availability of animal genetic resources influences both current and future quality of life and plays a crucial role in food security. The ratification of the Convention on Biological Diversity commits the Slovak Republic to the conservation of biodiversity, the sustainable use of its components (including animal genetic resources) and the fair and equitable sharing of benefits arising from their utilization. Additionally, the protection of animal genetic resources in agriculture aligns with the European Action Plan on Biodiversity, the UN FAO Global Strategy for the Management of Animal Genetic Resources, Regulation (EC) No 870/2004 establishing a Community programme on the conservation, characterization, collection and utilization of genetic resources and Regulation (EC) No 194/1998 concerning the breeding and stockfarming of animals, as well as the conservation of genetic resources.

It is vital to conserve and maintain animal genetic resources as a safeguard against threats, such as climate change, disease, social changes, genetic problems, selection errors and unexpected catastrophic events that could significantly impact biodiversity. Utilizing sperm, stem cells, bone marrow cells and oocytes from various sources offers a promising option for conserving endangered animal breeds. This process is preceded by *ex situ* collection and cell cultivation under *in vitro* conditions, testing for origin and quality and then cryopreserving the samples.

The submitted project will focus on the mapping, selection and genotyping of Slovak indigenous breeds, assessing semen quality, collecting and characterizing stem cells and evaluating the bacteriological profile of each sample. Once suitable individuals are identified, the collected samples will be stored in gene banks. Additionally, an

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"*ex situ in vivo*" collection of wild representatives of rabbits, poultry, sheep and hares will be established at the NPPC.

Cooperation with Hungary will focus on sharing animals, samples and methods to enhance gene conservation efficiency. The methodologies and instrumentation of the partner institutions complement each other, facilitating a more thorough evaluation of samples. The animal gene bank in the Slovak Republic will also store samples of Hungarian breeds and vice versa, ensuring that genetic material is available in both countries in the event of unforeseen circumstances.

The websites of the program and the project: <https://www.skhu.eu>, [www.nbgk.hu](http://www.nbgk.hu), [www.nppc.sk](http://www.nppc.sk)



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**Dr. István Bíró - Dr. László Zsilinszky: Formation and change of the structural system of breeding organization during the historical development of Hungarian livestock breeding - book review**

A new professional issue was published (in Hungarian) at the end of March 2024, Dr. István Bíró - Dr. László Zsilinszky: Formation and change of the structural system of breeding organization during the historical development of Hungarian livestock breeding (Tenyésztésszervezés szervezeti rendszerének kialakulása és változása a magyar állattenyésztés történeti fejlődése során; ISBN 978-615-01-9417-2). It has now fortunately been published with the support of the Ministry of Agriculture and the care of the DAGENE Association. The book's Introduction was written by András Gáspárdy, president of DAGENE. The publication may be of greater interest among agricultural education and research institutions, breeding organizations, and animal husbandry specialists.

It has been preserved in the works of writers, following the development history of our animal husbandry, that the main occupation of our conquering ancestors was animal husbandry. Upon their arrival in their new homeland, the Hungarian tribes possessed considerable livestock wealth. The natural characteristics of the Carpathian Basin provided ideal conditions for animal husbandry. Thus, this important sector remained a determining factor of economic life for centuries. The history and process of Hungarian livestock breeding spans a long period of time. Its development was strongly influenced by growing professional experience and organization.

Conscious animal husbandry is an extremely diverse and complex, but at the same time well-co-ordinated set of breeding activities. The genetic development work that foresees a quality animal product takes effect in the process of breeding organization of animal husbandry.

In the historical process of Hungarian livestock breeding, this book tries to trace the eras and development path of the documented breeding organization. It tries to show how long and arduous the professional path has been from the beginning during the conquest, through the development of conscious breeding, to modern selection systems, such as the domestic application of modern genome tests, to the present day. The self-sacrificing contribution and work of many excellent domestic professionals became the determinant of the success of each era. (See the personalities highlighted in the book!)

The centuries-old history and experiences of domestic livestock breeding clearly demonstrate that the eras that bring documentable development at the national level (in breeding programs, genetic progress, etc.) when the state, government bodies,

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and later the EU institutional systems with their specific (regulatory, financial, etc.) tools help and motivate the organization of breeding, the work of breeders and social organizations responsible for breeding. Excessive influence of the state, as it happened e.g. in the 1950s and 1960s, resulted in a decline in the same way as when government agencies left it alone, inadequately supporting the social actors of breeding (see the end of the 2000s!).

It is necessary to maintain and modernize the standard of domestic breeding, because even in the global market, the „market competition” has already intensified tremendously, both for domestic and foreign distribution of breeding animals, reproductive materials, etc., and in relation to the production of quality animal products. This is a big challenge for domestic breeding organizations designated and responsible for the genetic maintenance of breeds. Their market exposure and the compulsion to use the most advanced breeding tools was reinforced by „Regulation (EU) 2016/1012 of the European Parliament and Council”. It enabled intra-EU „transition” within the breed, i.e. any breed association under given „breeding authority” conditions, can enter as a competitor against the original, legally operating breeding organization of another EU member state.

At the same time, the EU legislative and institutional framework also provides the current framework and practical possibilities for the domestic breeding organisation (breeding programs, population genetics developments, etc.). Other professional civil organizations can collaborate creatively and „freely” and be part of international animal breeding programs. In the context of the above, the regulated breeding policy and the institutional system of the well-structured breeding organization can be connected to each other in the long term or even in the present, in order to successfully develop the domestic animal production and implement the necessary selection programs, in the case of all affected economic animal species.

It can also be stated that in today's „accelerated” world, the need in the Hungarian breeding organization for necessary use of results of international scientific development, such as genomics, artificial intelligence, etc., emphasizes even more the close co-operation of domestic breeding organizations that help each other. Furthermore, these stimulate the co-operation of domestic breeding organizations with the breeding authority, the agricultural administration and other bodies. The supportive government conditions, such as adequate budgetary support for the domestic breeding organization, the creation of tender opportunities for large investments (e.g. IT and special laboratory developments), etc., require continuous reinforcement. Preservation of the high standard of livestock breeding in Hungary is in the interest of the whole economy and stems from the traditions of our history in the Carpathian Basin!

András Gáspárdy