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### NEW ASPECTS IN RISK STATUS EVALUATION OF SMALL RUMINANT LOCAL BREEDS IN SERBIA

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Abstract: Several factors have led to a decline in autochthonous breeds, and the need for conservation programs based on risk assessment has increased. The aim of the study was to compare and validate two models for risk assessment of local small ruminant breeds in Serbia. The first model considered only the effective population size (Ne), while the alternative model included Ne, the number of females and sub-factors representing the influence of different elements. The results indicate an increasing trend in most breeds and populations over the last five years, with the exception of Tsigai and Chokan Tsigai, which showed a decrease. However, the increasing trend is insufficient, especially for Pirot, Karakachan, and Bardoka, as well as for the Domestic (Serbian) white goat, which is classified as critically endangered in both models. Vlashko vitoroga, Chokan tsigai, and the Balkan goat were at high risk. The models produced different results for the Krivovir, Sjenica, and Svrljig strains. In the first model, Krivovir was classified as potentially endangered, whereas Sjenica and Svrljig were not. In the second model, Krivovir was classified as highly and the other two strains as potentially endangered. These differences are due to the additional factors in the second model, which lead to a more comprehensive assessment for future risk assessments. Although the new model is appropriate for Serbia, it is important to test it on a variety of native species to increase confidence. Future conservation programs should also include genetic characterization and implement adaptive models to obtain more reliable conclusions.

**Key words:** sheep, goat, genetic resources, endangerment assessment, conservation

#### Introduction

The term animal genetic resources refers to the diverse pool of genetic material, such as genes and genetic traits, found in various domestic animal breeds and populations. Basically, these are local and autochthonous breeds that are specific to a particular area. These resources include inherent genetic variability that allows animals to adapt to different environmental conditions, exhibit different physical and physiological characteristics, and possess different production capabilities. Because of the unique genetic combinations, conservation and utilization of genetic resources is critical for several reasons, including biodiversity, adaptation and resilience, productivity, disease resistance, and adaptation to climate change.

Different specific environmental factors such as climate, geological and hydrological parameters, nutrition, animal migrations, and other factors have contributed to the formation of different ecotypes and breeds of sheep and goats that differ in morphological, reproductive, and productive characteristics. They represent a unique genetic combination and an important link in the agricultural biodiversity, tradition and cultural heritage of Serbia (Ružić Muslić et al., 2015). On the territory of Serbia, the representatives of autochthonous sheep breeds are Pramenka (Zackel) with its differentiated strains (Cekic et al., 2018) and Tsigai (Tsigai and Chokan Tsigai), and the representatives of goats are Balkan goat and Domestic (Serbian) white goat (Maksimović et al., 2023). Due to various factors (political, demographic, economic, etc.), autochthonous breeds were displaced by imported, conditionally more productive breeds intented for intensive systems, which affected the decreasing number of animals of local breeds. *Reist-Marti et al.* (2003) claimed that the introduction of imported (exotic) genotypes threatened at least 1000 breeds with extinction worldwide, representing a major loss of genetic diversity. For sustainable management of animal genetic resources in a sustainable way, their monitoring and characterization must be introduced (ERFP, 2021). As the size of local breed populations decreases, the need for risk assessment increases.

Assessment of livestock risk status is a key component of early warning and response systems in countries (*Verrier et al., 2015*) and has been studied in many countries (*Polak et al., 2021*) using a variety of methods (*Urošević et al., 2022*). Initially, assessments were based on total population size and the proportion of matings in the pure breed (*Scherf, 2000*), but later methods incorporated other factors, such as demographic, genetic, and socioeconomic factors (*FAO, 2007*), as well as the time required to reach a critical population size (*Gandini et al., 2005*). Furthermore, *Alderson (2009)* claimed that in addition to the above factors, the occurrence of the same breed in many countries, as well as anthropogenic, climatic, and epidemiological factors should be considered when assessing risk status. Also, FAO (2013) published guidelines for breed conservation based on total population size, breeding population size, and reproductive capacity of species, and established the Domestic Animal Diversity Information System (DAD-IS) and the European Farm Animal Biodiversity Information System (EFABIS) to assess risk status.

Nowadays, the methods for assessing risk status and their specific factors are country-dependent, and most of them focus on population size and the measures needed to reduce inbreeding rates and maintain heterozygosity (*Notter*, 1999). Although there is a lack of harmonization of procedures, most conservation programs classify breeds according to extinction risk based on their endangerment status (*Alderson*, 2009). Based on this fact, the aim of this study is to evaluate the endangerment status of local breeds of small ruminants in Serbia by using two models, comparing and validating their results.

### **Materials and Methods**

Data on population size, and number of breeding animals (male and female) under control of productive traits were taken from databases and annual reports on activities and results of control of breeding programs implementation in 2022 of the two main breeding organizations for small ruminants in the Republic of Serbia - Institute for Animal Husbandry, Belgrade (for Central Serbia) and Department of Animal Science, Faculty of Agriculture, University of Novi Sad (for Vojvodina region), as well as DAD-IS database of *FAO* (2021).

Two approaches were used to assess the risk status of local breeds in Serbia. The first one corresponds to the official model of the Ministry of Agriculture, Forestry and Water Management described in the Decree on the List of Domestic Animal Genetic Reserves, the Method of Conservation of Domestic Animal Genetic Reserves and the List of Native Domestic Animal Breeds and Endangered Native Breeds (*Official Gazette 30/22*), while the second one represents the model described and proposed by *Polak et al. (2021)*. In both, effective population size (Ne) was used as the main parameter for representing genetic diversity and inbreeding rates in animal populations

The official model focused on risk assessment using only effective population size, using the formula:  $Ne = 4Nm \times Nf/(Nm+Nf)$ , where Nm is the number of breeding males and Nf is the number of breeding females. In terms of the degree of endangerment, populations are divided into four groups: critically endangered (Ne  $\leq$  50), highly endangered (Ne  $\geq$  50 and N  $\leq$  200), potentially endangered (Ne  $\geq$  200 and N  $\leq$  1000), and not endangered (N  $\geq$  1000).

The alternative model used the correction of Ne by multiplying it by a factor of 0.7 (Ne<sub>c</sub>), assuming random selection of males and females for breeding (Santiago and Caballero, 1995). In addition to Ne<sub>c</sub>, the method considered additional factors and included the total number of breeding females (L) and a complex factor (SSF) composed of six elements: geographic concentration within a country (GC), demographic trend over the past five years (DT), cultural value of the breed (VC), control of ancestry (PC), presence of ex situ conservation (EX), and anthropogenic factors that include breeder age, implementation of existing conservation programs, and potential financial support for breed conservation (AF). To mitigate the potential effects of subjectivity and its potential influence on results, the SSF factor is divided by 1/2, reducing the likelihood of estimation error. If L is less than 150, breed is classified as critically endangered, regardless of the final outcome. Further descriptive details of the parameters and subfactors are published in Polak et al. (2021). Each of the listed parameters was defined by the thresholds and scoring values listed in Table 1, and the following formula was used to calculate the risk status (RS) of each strain/breed:  $RS = (L + Ne_c + 0.5SSF)/3$ . Based on the RS value, populations are sorted as follows:  $\leq 1$  - critical;  $> 1 \leq 2$  highly endangered breed required action; > 2 < 3 - potentially endangered breed requiring monitoring; > 3 - not at risk.

<b>D</b> (	Score value								
Parameter	0 0.5		1	2	3				
Total number of females L	< 150	/	151 - 1000	1001 - 6000	6001 - 20000				
Corrected Effective population size Ne <sub>c</sub>	≤ 50	/	50 - 200	201 - 1000	> 1000				
Geographical concentration	$\leq 25\%$	25 - 75%	>75%	/	/				
Demographic trend	Down	Stable	Upward	/	/				
Cultural and historical value	Present	Small value	No value	/	/				
Parentage control	Lacking	Existing to a small extent	Existing	/	/				
<i>Ex situ</i> conservation	Lacking	Existing to a small extent	Existing	/	/				
Anthropogenic factors	Lacking	Existing to a small extent	Existing	/	/				

Table 1. Scoring range regarding the factors and sub factors for risk evaluation

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#### **Results and Discussion**

Regarding the number of animals in the last five years, Table 2 shows that there is an upward trend in most populations, except for Chokan Tsigai and Tsigai, where the number of animals is lower in 2022 compared to 2021. Although for most observed populations this trend is promising, it is still not satisfactory. This is of particular concern for breeds that are not transboundary and are not regionally distributed, such as the Pirot strain of Pramenka and the Domestic (Serbian) white goat. In these populations, intervention by importing new, unrelated animals is not possible, and the risk of inbreeding is higher. In a small population, inbreeding increases because after one generation each individual is related to some degree to every other individual (*Meuwissen, 2009*), which leads linearly to a decrease in genetic variance (*Falconer and Mackay, 1999*).

	YEAR								
STRAIN / BREED	2018	2019	2020	2021	2022				
SHEEP									
Pirot strain	129	149	197	195	302				
Karakachan strain	165	176	186	225	252				
Krivovir strain	816	1,103	1,118	1,600	2,068				
Bardoka strain	117	107	142	144	207				
Sjenica strain	74,604	105,573	139,319	176,534	199,571				
Svrljig strain	14,807	18,166	19,585	25,857	29,285				
Lipa strain	941	1,280	1,636	2,102	2,866				
Vlashko vitoroga (Racka)	331	492	1,060	1,131	1,150				
Chokan tsigai	1,098	1,834	2,092	2,505	2,485				
Tsigai	5,580	5,580 5,613 5,161		4,362	4,227				
GOAT									
Balkan goat	427	590	791	921	1.153				
Domestic (Serbian) white	138	152	168	147	201				

The goal of comparing the two methods was to evaluate various factors that classify a particular breed as being in danger of extinction. The results presented in Table 3 include the risk assessment according to the official model, considering all four endangerment categories. Thus, in the group of critically endangered populations, Pirot, Karakachan and Bardoka strain, Vlashko vitoroga and Chokan tsigai are at high risk, while Krivovir and Lipa strain and Tsigai are at potential risk. On the other hand, the most numerous populations of local sheep breeds (Sjenica and Svrljig) are not threatened with extinction. In the case of goats, both breeds are threatened with extinction, while the Serbian (domestic) white is critical and the Balkan goat is highly endangered. As mentioned above, this model is based on Ne, one of the most important parameters in monitoring local breed populations (*Verrier et al., 2015*), as it allows predicting changes in genetic variance (*Wright, 1931*).

STRAIN / BREEDS	Number of females	Number of males	Ne	Risk status					
SHEEP									
Pirot strain	290	12	46.09	critically endangered					
Karakachan strain	240	12	45.71	critically endangered					
Krivovir strain	1.980	88	337.02	potentially endangered					
Bardoka strain	198	9	34.43	critically endangered					
Sjenica strain	194,342	5,229	20,367.98	non endangered					
Svrljig strain	28,367	918	3,556.89	non endangered					
Lipa strain	2,780	86	333.68	potentially endangered					
Vlashko vitoroga (Racka)	1,102	48	183.99	highly endangered					
Chokan tsigai	2,445	40	157.42	highly endangered					
Tsigai	4,059	168	645.29	potentially endangered					
GOAT									
Balkan goat	1,115	38	146.99	highly endangered					
Domestic (Serbian) white	191	10	38.01	critically endangered					

Table 3. Risk status of small ruminants local breeds according to official model

Using the model proposed by *Polak et al. (2021)*, the results regarding risk assessment of local strains and breeds are similar to the previous model, and the Pirot, Kakarachan, and Bardoka strains and the Domestic (Serbian) white goat are listed in a group of critically endangered populations, while Lipa, Vlashko Vitoroga, Chokan Tsigai and Tsigai, and the Balkan goat are highly endangered populations requiring action, as listed in Table 4. This similarity may indicate that

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the newly proposed model can be effectively used to assess the risk status of small ruminants under the conditions prevailing in the Republic of Serbia. The main difference between these two models is with respect to the strains Krivovir, Sjenica and Svrljig. While the results of the official model showed that the Krivovir strain is potentially endangered, and the other two strains are stable, non endangered, the results of the alternative model showed that Krivovir is highly endangered, while Sjenica and Svrljig are potentially endangered, and that surveillance is still needed. This discrepancy is the result of the use of additional subfactors detailed in terms of demographic, cultural, anthropogenic, and conservation aspects that enhance the risk assessment approach. The results suggest that these factors have a real and strong influence on the sustainability of a strain, breed or population.

STRAIN /	$L^1$	Score		Score Ne <sub>c</sub> <sup>4</sup>	Sub factors					<b>D</b> C <sup>12</sup>	Risk		
BREEDS	L	$L^2$	Ne <sub>c</sub> <sup>3</sup>		GC <sup>5</sup>	DT <sup>6</sup>	VC <sup>7</sup>	PC <sup>8</sup>	EX <sup>9</sup>	AF <sup>10</sup>	SSF <sup>11</sup>	RS <sup>12</sup>	status <sup>1</sup>
SHEEP													
Pirot strain	290	1.00	32.26	0.00	0.50	0.00	1.00	0.50	1.00	1.00	4.00	1.00	CE
Karakachan strain	240	1.00	32.00	0.00	0.50	0.00	1.00	0.50	1.00	1.00	4.00	1.00	CE
Krivovir strain	1.980	2.00	235.91	2.00	1.00	0.00	1.00	0.50	0.00	1.00	3.50	1.92	HE
Bardoka	198	1.00	24.10	0.00	0.50	0.00	1.00	0.50	1.00	1.00	4.00	1.00	CE
Sjenica strain	194,342	3.00	14,257.58	3.00	1.00	1.00	1.00	0.50	1.00	0.50	5.00	2.83	PE
Svrljig strain	28,367	3.00	2,489.83	3.00	1.00	0.00	1.00	0.50	0.50	0.50	3.50	2.58	PE
Lipa strain	2,780	2.00	233.57	2.00	1.00	0.50	1.00	0.50	0.50	1.00	4.50	2.08	PE
Vlashko vitoroga (Racka)	1,115	2.00	128.79	1.00	1.00	0.50	1.00	0.50	0.50	1.00	4.50	1.75	HE
Chokan tsigai	2,453	2.00	110.20	1.00	1.00	0.50	1.00	0.50	1.00	1.00	5.00	1.83	HE
Tsigai	4,097	2.00	451.70	2.00	1.00	0.50	1.00	0.50	1.00	1.00	5.00	2.17	PE
GOAT													
Balkan goat	1,115	2.00	102.89	1.00	0.50	0.00	1.00	0.00	1.00	1.00	3.50	1.58	HE
Domestic (Serbian) white	191	1.00	26.61	0.00	0.50	0.00	1.00	0.50	1.00	1.00	4.00	1.00	CE

Table 4. Risk status of small ruminants local breeds according to number of females, corrected effective population size and additional sub factors

 $^1L$  - Number of breeding females;  $^2Score$  value regarding the L;  $^3Ne_c$  - Effective population size corrected for uncontrolled breeding;  $^4Score$  value for Nec;  $^5GC$ - sub factor of geographical

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concentration; <sup>6</sup>DT - sub factor for trend of demography; <sup>7</sup>VC - sub factor representing the cultural value of breed/strain; <sup>8</sup>PC - sub factor representing the value of parentage control; <sup>9</sup>EX - value regarding to existence of *ex situ* conservation; <sup>10</sup>AF - value of anthropogenic factors; <sup>11</sup>SSF - Total value of sub factors; <sup>12</sup>RS - risk status; <sup>13</sup>CE - critically endangered, HE - highly endangered breed requiring action, PE - potentially endangered breed, requiring monitoring.

One of the most important factors contributing to the increase in the risk of extinction is anthropogenic factors. For conservation of local breeds, the sociopolitical context of humans must be understood as it must have some impact on the livelihood of breeders (*Gizaw et al., 2008*). Conservation of local breeds should be cost-effective and based on a multi-criteria decision (*Wainwright et al., 2019*). *Meuwissen (2009)* claims that the best strategy to conserve an endangered breed is to make it profitable. In the Republic of Serbia, there are governmental measures in the form of subsidies whose main goal is to achieve profitability and economic efficiency of genetic resources (*Nastić et al., 2020*). However, they cannot always cover the increasing production costs. Another critical factor is the low awareness of breeders about the importance of local breeds.

In addition, demographic trends over the last five years are an important factor that can be used to objectively assess population trends. The main areas for breeding local sheep and goat populations are rural areas, and with depopulation, the number of livestock in general is decreasing, as well as the number of genetic resources.

The cultural value of the breed and its historical significance are important factors that should be considered in risk assessment. These breeds have great potential in the artisanal processing of milk, meat, and wool into traditional products that have a strong influence on folklore, gastronomy, tourism and religious tradition. In other words, the loss of local breeds has an impact on the loss of cultural value and the loss of historic landscapes (*Meuwissen, 2009*).

A model based on a multifactor analysis is certainly more comprehensive than one that uses only Ne as a risk assessing element. In addition, the use of a larger number of indicators leads to a more continuous view of the degree of risk. Obtaining the information, defining the indicators, and establishing the score values is also relatively straightforward, as the two main breeding organizations have information to track breeds in terms of population size and structure, as well as productive traits. In addition, combining several different indicators can minimize the possibility of undermining the purpose of the score. In other words, when the number of indicators in the model is increased, the reliability of the assessment at risk increases and the possibility of error is reduced.

However, it should be noted that while this model works for small ruminant breeds in Serbia, its suitability should be investigated for other domestic species. To ensure even greater reliability, future conservation programs should include, in addition to these factors, the aspect of genetic conservation, the main objective of which would be to maintain breed diversity. The models could be modified according to the availability of new information on novel indicators or subfactors so that the information is wholesome for their calculations.

As stated by *Ružić-Muslić et al. (2021)*, the concept of sustainable use of genetic resources consists in their identification, description, development and monitoring, leading to their conservation. To ensure efficient management of genetic resources, it is important to maintain breed registers, conduct genetic evaluations, promote controlled breeding initiatives, and support sustainable breeding practices. These measures are essential to prevent genetic erosion and improve the overall quality of small ruminant populations. The models used to assess vulnerability must be able to evolve as new information is obtained and as legislation and objectives change.

#### Conclusion

Sustainable use of genetic resources requires their identification and description, development and monitoring, and consequently methods for their conservation. One of the ways to perform monitoring and propose development measures is to assess the risk of extinction. In this study, two risk assessment models were used and their results are compared. In the majority of the observed populations, there is an increasing trend in the number of animals, with the exception of Tsigai and Chokan Tsigai, which are decreasing. However, in the populations of Pirot, Karakachan, Bardoka (sheep) and Domestic (Serbian) white goat, which are classified as critically endangered in both risk status assessment models, the number of animals is insufficient. Although the results regarding the risk status of both models are similar, the alternative model provides a broader perspective as it takes into account demographic, sociocultural, geographic, anthropogenic, and conservation factors. This model was proposed for Poland, but the results show that it can also be used to assess local breeds in Serbia. Further research is needed to assess the risk status for other species of livestock genetic resources. To ensure greater reliability of results, future work should also focus on aspects of genetic characterization, with the main goal of obtaining meaningful information that can be implemented in conservation programs.

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