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# RESEARCH ON DIVERSITY, UTILIZATION AND PRODUCTION QUALITY OF LOCAL BREEDS IN SLOVAKIA

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Scientific review

## SUMMARY

*State of the research in the area of utilization of local breeds in Slovakia provides sufficient information for the farmers and breeders associations either on level of diversity based even on pedigree or molecular information as well as the information on molecular composition of milk and meat for animals selection to produce specific quality products. With research and development applied we aim not only to protect something ancient, historical, but also provide progress to increase profitability with ensured safety and sustainability. Present research in area of local breeds utilization is oriented on study of functional traits in cattle as well as sport performance traits in warm blood horses.*

*Key-words: degree of endangerment, genetic diversity, livestock, local breeds*

## INTRODUCTION

In the last 40 years, production ability improvement of farm animals was mainly assigned (85%) to the genetic and animal breeding. The main challenge, pushing the research forward, is that the World needs more food. According to conclusion of the World food summit held in Rome, the agricultural production will increase by 70% till 2050. The farmers say that today's farm management isn't improving as fast as animal genetics. In front of us is the responsibility to show consumer future of the farms in form as it was made for example with computers. The responsibility on the other hand means that with afforded opportunities there are many expectations facing us, as to make agriculture attractive and to increase the profitability and ensure the sustainability. Using local breeds we try to tell our story that farm life is attractive, providing it to consumers as a brand. With research and development applied we aim not only to protect something ancient, historical, but also provide progress to increase profitability with ensured safety and sustainability.

With increased demand for animal origin food, some branches became very intensive, like milk production, pork production, poultry production using of specific breeds or hybrids even more. Traditionally bred local breeds became marginal. After 40 years of breeding for increased production those modern breeds miss the story, they lack adaptability, immunity, multi-purposity

and robustness as known from formerly kept breeds. These are the opportunities provided by local breeds.

## STATE OF THE BREEDS ENDANGERMENT IN SLOVAKIA

Breeds of horses, cattle, goat, sheep and poultry (Table 1) belong to the farm animal genetic resources in Slovakia (AnGR). Slovak spotted and Slovak Pinzgau whose origin is composite of autochthonous Carpathian Red (extinct) and Carpathian grey (extinct) from 17<sup>th</sup> to 18<sup>th</sup> century as well as Swiss Simmental and Austrian Pinzgau from 19<sup>th</sup> century common in Austro-Hungarian Empire belong to the main cattle breeds of national interest. Wallachian sheep and Hucul horse became present from Wallachian migration to Area of today's Slovakia. Even Original Wallachian sheep was replaced by Wallachian improved breed. Today there are activities to re-establish stock of original Wallachian sheep based on back-cross of negative variants. Besides Hucul, there are other horse breeds which were traditionally bred for purposes of Austro-Hungarian army: Furiosso, Nonius or aristocracy: Arab, Shagya-Arab, Lipizan, English thoroughbred. The heavy draught horse represents Norik of Muran. Experimental breeding of warm blood horses

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resulted in Slovak Sport Pony or Slovak Warm Blood horse. Breeder's interests re-started the Brown Short Hair goat breeding whereas national Suchovska goose or Oravka chicken hardly survives.

**Table 1. Inventory of Farm Animal Genetic Resources in Slovakia (Hetényi et al., 2006)**

Species	Breeds			Total
	Local	New import	Long time import	
Cattle	2	8	1	11
Sheep	4	4	4	12
Goat	2	1	-	3
Pig	4	-	4	8
Horse	9	-	5	14
Fur animals	10	-	1	11
Rabbit	12	-	39	51
Poultry chicken	7	-	24	31
Duck	2	-	3	5
Goose	4	-	2	6
Turkey	1	-	3	4
Quail	1	-	3	4

## METHODS OF EVALUATION OF DEGREE OF ENDANGERMENT

### Pedigree based evaluation

Kadlečík et al. (2007, 2011) evaluated complexity of pedigrees and equivalent parameters in purebred Slovak Pinzgau population and estimated intensity of inbreeding. From 1994 the population has been recognized as endangered. Hazuchová et al. (2010) evaluated the diversity of Pinzgau population based on pedigree information of active AI sires, estimated inbreeding levels and characteristics based on probability of gene origin. Lower pedigree completeness as well as unequal founder contribution was observed due to use of only several sires, but with positive tendency in inbreeding decrease. Hazuchová et al. (2012, 2013) evaluated genetic diversity of Slovak spotted bulls using parameters based on gene origin probability. Even with population not endangered signs of diversity loss were observed due to decrease in effective number of founders in reference population as well as overall decrease of genetic diversity as the consequence of unequal contribution of founders. Diversity of cattle breeds was also evaluated by Kadlečík et al. (2013). The whole analysed populations consisted of (reference populations in brackets) 274,756 (94,357) Holstein, 109,686 (36,949) Slovak spotted and 9,756 (2,501) Slovak Pinzgau cattle. Indices of pedigrees completeness differed by breeds but in the 5<sup>th</sup> generation their values were 18.4% in Slovak Pinzgau, 56% in Slovak Spotted breed and 63.8% in Holstein. Generation interval of Slovak Pinzgau was 7.2 years, tending to be shorter as the population size

was bigger. There was 30% of inbred Slovak Pinzgau, 42.8% in Slovak Spotted and 83% in Holstein cows and sires in the reference populations. The average inbreeding coefficient ranged from 0.36% for the Slovak Spotted to 1.32% for the Holstein. The highest average individual increase of inbreeding  $\Delta F_i=0.29\%$  was found for the Holstein and the average individual relatedness coefficient  $AR=0.8\%$  in Slovak Spotted cattle. Inbreeding trends in the whole reference populations as well as purebred animals were positive with increasing average values by animal birth years since 1990. Pavlík et al. (2014) performed diversity joint genealogical analysis evaluation of Pinzgau cattle populations in Slovakia and Austria and found minimum differences between respective populations. The same authors (2014) evaluated basic measures of diversity in Slovak Holstein (H) and Red-Holstein (R) populations by the pedigree analysis. Results presented higher inbreeding level in H than in R including higher increase in inbreeding. On the other hand diversity in R population is more significantly reduced by bottleneck and genetic drift.

Pjontek et al. (2010) analysed genetic diversity of endangered thoroughbred Arab horse population based on description probability of identity by gene descent and origin. They have observed that the high number of founders registered in the studbook would suggest the existence of a large genetic basis for a stallion's selection program. However, historical constraints and the breeding policy carried out by the breeders have reduced the available genetic variability as reflected in small values computed for parameters such as the effective number of founders and ancestors. Pjontek et al. (2012) conducted an analysis of the genetic diversity in four endangered horse populations bred in Slovakia, describing parameters on the probability of identity by descent and gene origin. The whole analysed populations consisted of (reference populations in brackets) 656 (158) Hucul horses, 2052 (162) Lipizan horses, 1951 (171) Shagya Arabian horses and 220 (42) Slovak Sport Ponies. The equivalent complete generations ranged from 4.93, for the Slovak Sport Pony, to 10.25, for the Lipizan horses. The average value of inbreeding ranged from 2.67%, for the Slovak Sport Pony, to 6.26%, for the Hucul. The average relationship coefficients were from 3.08%, for the Shagya Arabians, to 9.34%, for the Huculs. Individual increases in inbreeding ranged from 0.43%, for the Lipizans, to 1.06%, for the Huculs, while the realised effective sizes were from 117.14 to 47.67 animals. The evaluated populations derived from 80 to 499 founders. The effective number of founders ranged from 26 to 160, while the effective number of ancestors from 7 to 32. Kadlečík et al. (2012), Kadlečík and Kasarda (2014) assessed genetic diversity of Slovak Sport Pony nucleus based on pedigree information. Pavlík et al. (2014) evaluated the breeding population of Thoroughbreds in Slovakia using pedigree analysis. The examined population consisted of 123 mares and 10 stallions registered in Slovak Stud Book. The average inbreeding coefficient was 0.86% in the whole

reference population (133 individuals) and increase in inbreeding per generation was 0.17%, in stallions 0.25%. The number of founders in the investigated population was 949. The effective number of founders was 202, while the effective number of ancestors was 67. These results point out unbalanced contribution of founders and ancestors into reference population as well as the bottleneck effect occurrence.

### Molecular based evaluation

Židek and Kasarda (2010) provided information on the genetic structure in Pinzgau cattle in Slovakia based on polymorphism of 61 monovalent sera. Genetic distance between 777 animals has been computed. Obtained genetic distances were pooled according to relationship coefficient  $R_{XY}$ . Average genetic distance decreased from 0.225 (group with  $R_{XY}$  between 0 and 0.125) to 0.147 (group with  $R_{XY}$  between 0.375 and 0.5). Šidlová et al. (2013), Šidlová et al. (2014 a, b) evaluated genetic diversity of Slovak Pinzgau cattle using 8 microsatellite markers (TGLA122, CSSM66, TGLA227, ILST006, CSR60, ETH3, BM1824, SPS115). Microsatellites were highly polymorphic with a mean number of 11 alleles (ranging from 9 to 16 per locus) and total number of 88 alleles. High level of polymorphism confirms also the average value of PIC (0.7662). The overall average of observed and expected heterozygosity has reached similar values (0.7927 and 0.7980), but the differences are noticeable at each locus separately. Židek et al. (2014) used molecular information on microsatellites to model case of missing pedigree information, when other methods can be used for traceability of animal's origin. They concluded that genetic diversity written in genetic data is holding relatively useful information to identify animals originated from individual countries. Kasarda et al. (2014a, b), Šidlová et al. (2014a) evaluated the level of SNP polymorphisms and described the basic characteristic of the analysed population genotyped using the BovineSNP50 genotyping array. In total 19 purebred Pinzgau bulls were successfully genotyped with Illumina BovineSNP50 BeadChip (98.96% of SNPs) with average call rate 0.995. Genotyping results from 54,906 SNPs revealed that 43,120 SNPs (78.96%) were polymorphic with average minor allele frequency  $0.273 \pm 0.133$ . Within 43,120 SNPs genotyped, 98.19% were autosomal, with 776 polymorphic SNP on chromosome X and only one on chromosome Y. The average values of the observed and expected heterozygosity across polymorphic loci were  $0.375 \pm 0.157$  and  $0.362 \pm 0.126$ , respectively. Sufficient proportion of heterozygotes indicated the value of  $F_{IS}$  ( $0.037 \pm 0.031$ ). Šidlová et al. (2014b, 2014c) derived inbreeding coefficient from runs of homozygosity. The highest level of autozygosity ( $F_{ROH1} = 10.88\%$ ) as well as the longest ROH segments in total (271.84 Mb) has been observed in Slovak bull Norfolk having Austrian origin. Austrian bull Nero had the highest number of ROH (80) and the second top inbreeding (9.63%). Carlo, Slovak bull of Canadian origin

had the lowest number of ROH, length of ROH segments and inbreeding level. Performing analyses with ROH of different lengths here allows estimation of the distance of the current population from the base population, hence provides information on inbreeding age. Previous results reported from other study on Pinzgau breed from Austria ( $F_{ROH1} = 0.069$ ) shows higher inbreeding levels than those found in this study on Slovak Pinzgau ( $F_{ROH1} = 0.0519$ ). It is also noticeable that bulls with Austrian origin have overall higher  $F_{ROH}$  levels.

### DIVERSITY OF PRODUCTION TRAITS

Trakovická et al. (2013a) and Moravčíková et al. (2013) analysed in total 296 blood samples of Slovak Spotted and 85 hair roots samples of Pinzgau cows to verify the associations of polymorphisms in bovine *LEP* and *LEPR* genes with production and reproduction traits in Slovak Spotted and Pinzgau cows. Long-life production: milk, protein, and fat yield and reproduction traits: age at first calving, calving interval, days open, and insemination interval were evaluated. Trakovická et al. (2013b) analysed genetic diversity in population of Slovak Spotted cattle based on *Pit-1/Hinfl* polymorphism. Moravčíková et al. (2012a) analysed association of bovine growth hormone gene polymorphism with milk performance traits in Slovak Spotted cows and Moravčíková et al. (2012b) studied polymorphism in the intron region of the leptin gene on bovine chromosome in relation to evaluation of genetic diversity. Moravčíková et al. (2013) identified SNPs of leptin (*LEP*), leptin receptor (*LEPR*) and growth hormone (*GH*) genes in order to analyse genetic diversity of Slovak Spotted cattle and evaluate their effect on production traits. Meluš et al. (2008a,b) analysed *SCD1* gene polymorphism (T878C) in the Slovak Pinzgau steers in relation to the haematological parameters. Miluchová et al. (2013) identified A1 variant of bovine beta casein which involves ischemic heart disease and diabetes mellitus in human. The digestion of A1 beta casein can result in the production of bioactive beta casomorphin-7 (BCM-7) whereas this is not the case with A2. In the total population of cattle homozygotes A2A2 (0.5405) were the most frequent, while homozygotes A1A1 (0.1261) were the least frequent ones. This suggests a superiority of allele A2 (0.7072) which does not produce BCM-7, and thus is safe for human consumption. The expected homozygosity for gene *CSN2* was in the population stating a slight increase in homozygosity (0.5858). This caused a slight decrease in the level of possible variability realization (41.80%), which corresponds to the effective number of alleles (1.7071). Miluchová et al. (2014) studied genetic structure of five candidate genes for milk production in Slovak Pinzgau breed. A total of 86 mothers of bulls of Slovak Pinzgau cattle were used in this study. Slovak Pinzgau cattle exhibited the high values of heterozygosity, polymorphism information content, effective number of alleles and level of possible variability realization for genes *CSN2*, *CSN3* and *LALBA*.

In opposite, high values of homozygosity were observed for genes *CSN1S1* and *LGB*. Meat tenderness is one of the major characteristic qualities of beef not only for consumers but for breeders of beef cattle too. Selection of cattle focussed on an increment of meat tenderness is complicated because this trait has large variability not only between different breeds but between individuals of equal breed too. Similarly a measurement of meat tenderness is expensive because it is done after slaughter of animal and ageing of meat *post mortem*. Therefore a several methods are developed and made possible to increase meat tenderness. However variance still exists in values of meat tenderness caused by distinctness genetic base of animal. The most significant candidate genes (*CAPN1*, *CAST*) coding formation of the calpains – calpastatin proteolytic system, exercising an influence on tenderness was described by using molecular genetics methods. The single nucleotide polymorphisms (SNPs) in these genes were used by Gábor et al. (2010) to design genetic marker panels applying commercially available test. Gábor et al. (2012) analysed the population of 113 animals of Slovak Simmental (42 bulls and 71 cows) for the missense mutation resulting in SNP polymorphism in exon 3. The SNP in *CAST* gene (c.283 C>T) was detected by PCR-RFLP method with restriction endonuclease *MspI*. The following frequency of alleles and genotypes for the SNP c.283 C>T of the *CAST* gene were detected in the analysed population of Slovak Simmental cattle. Frequencies of favourable C allele were 0.6460 whereas of genotypes were 0.4336 (genotype CC), 0.4248 (genotype CT) and 0.1416 (genotype TT).

## CONCLUSION

State of the research in the utilization area of local breeds in Slovakia provides sufficient information for the farmers and breeders associations either on level of diversity based even on pedigree or molecular information as well as the information on molecular composition of milk and meat for selection of animals to produce products of specific quality. Provided information present missing innovation necessary for farmers to stay sustainable. The studied populations were evaluated in complex not only quantitative but also qualitative measures allowing global view on future performance of local breeds and populations. The present research in area of local breeds utilization is oriented on study of functional traits in cattle as well as sport performance traits in warm blood horses. Further research in area on genetic diversity will be molecularly based to get detailed view on genetic structures.

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