EVALUATION OF GENETIC DIVERSITY IN SELECTED BEEF BREEDS

Kristína LEHOCKÁ*, Radovan KASARDA, Barbora OLŠANSKÁ, Nina MORAVČÍKOVÁ

Department of Animal Genetics and Breeding Biology, Slovak University of Agriculture in Nitra, Slovakia
*Corresponding author: tina.lehocka@gmail.com

ABSTRACT

The aim of the study was to estimate genetic drift and gene flow related to population structure and genetic diversity in selected beef cattle. For the evaluation of the genetic drift and gene flow among analysed populations, the Bayesian Population Structure Analysis and software Treemix were used. The genetic analysis included two cattle breeds bred in Slovakia (Charolais and Limousine). In addition to the Limousine and Charolais breeds, other beef cattle (Angus N = 90, Belgian Blue N = 4, Blonde d'Aquitaine N= 5, Hereford N = 98 and Red Angus N = 15) were analysed. The 50k Bead chip was used; the dataset consisted of 34,834 SNPs. To avoid detection bias, SNPs with high linkage disequilibrium (r^2 = 0.05) were pruned from the database; the final data set consisted of 296 animals and 2,539 SNP markers. Our results reflected four modes of gene flow between Angus, Red Angus, Charolais, Limousine and Hereford. Analysed breeds were not confirmed to influence genetic make-up of Belgian Blue and Blonde d'Aquitaine populations. All migration edges reached weight values below 0.2. The only two migration edges higher in weight was observed, first between the ancestor of Limousine breed into Blond d'Aquitaine, and second among historical ancestor of Hereford breed into Red Angus. Our results reflect that the donor population has made a significant contribution to the recipient population.

Keywords: *beef, Charolais, gene flow, Limousine, Treemix.*

INTRODUCTION

Genetic diversity is an important indicator used in improvement and conservation programs both in cattle and in other livestock populations (Hlongwane et al., 2020). Management of genetic resources in livestock populations is an important measure for maintaining the production of animal products (Makanjuola et al., 2020). Charolais and Limousine are the original French breeds of cattle, whose breeding history has 30 years tradition in Slovakia. Both breeds that were imported to Slovakia in 1990 (Kadlečík et al., 2016) belong today to the most popular breeds for use in utility crossbreeding (Association of Slovak Spotted cattle breeders -

Cooperative, 2020). Genetic diversity is an indicator of interest for both breeders and population geneticists as its status and level of variability reflect developmental processes such as adaptation, selection, gene flow and drift among populations (Goszczynski et al., 2014; Bohórquez et al., 2020). Genetic drift represents a random change in the allele frequency of an existing gene variant in a population as a result of a random sampling of organisms (Masel, 2011; Merilä, 2014). Gene flow plays a key role in the transfer of genetic diversity among populations (Bolnick and Nosil, 2007). Groeneveld et al. (2010) stated that different types of markers could be used to evaluate the state of genetic diversity or population structure, and in recent years single nucleotide polymorphism (SNP) analyses have been used in research. SNP markers are more commonly used in the analysis of genetic diversity, represent a more advantageous method due to the higher level of resolution when compared to a microsatellite marker (Vargas et al., 2016).

The aim of this study was to evaluate population structure and genetic diversity based on the estimation of gene flow and genetic drift in beef breeds bred in Slovakia.

MATERIAL AND METHODS

The SNP database consisted of 296 animals from seven cattle breeds, including two Slovak populations of Charolais (N= 67) and Limousine (N=17) cattle. The other breeds represented publicly available data of Angus (N = 90), Belgian Blue (N = 4), Blonde d'Aquitaine (N = 5), Hereford (N = 98) and Red Angus (N = 15) cattle. For Charolais and Limousine breeds, genomic DNA was extracted from the hair roots and subsequently genotyped in a commercial lab using the International Beef and Dairy (IDB) chip. The genotypic data of other breeds (Angus, Belgian Blue, Blonde d' Aquitaine, Hereford and Red Angus) were obtained using web-based data archive (McTavish et al., 2013). Quality control was performed for seven beef breeds using the PLINK v1.9 program (Chang et al., 2015), according to Kukučková et al. (2017). SNPs that reached higher linkage disequilibrium (LD) were pruned out from the database. It has been shown that pruning of SNPs with high LD counteracts the effect of the ascertainment bias and consequently makes a meaningful comparison between breeds (Kijas et al. 2009). Total of 2,539 SNPs remained for further analysis. Population structure among beef populations was estimated by Bayesian Population Structure Analysis (BAPS) version 6.0 (Corander and Tang, 2007). According to Kukučková et al. (2017) were set the number of cluster K 7-50 and 1000 simulations from the posterior allele frequencies. For recognition of genetic drift and gene flow among populations were used the Treemix program (Pickrell and Pritchard, 2012). First, a phylogenetic tree of evaluated bovine populations was created, which was based on maximum probability, and then migration edges (m = 8) were added to the generated graph (Upadhyay et al., 2019). All graphs were visualised using the statistical software R (R Core Team, 2014).

RESULT AND DISCUSSION

Based on our results, it is possible to see the occurrence of gene flow between populations (Figure 1). Arrows point on the specific populations in the graph, expressing the given degree of admixture of individual populations. Tang et al. (2009) reported that a typical population, in addition to own primary resources, is mostly made up of small portions of gene flow from other populations. The observed level of admixture in this study was expressed in the graph as arrows between populations, with the corresponding numbers being the result of random generation from a uniform distribution (Kukučková et al., 2017). Our results reflect four modes of gene flow between Angus (cluster 1), Red Angus (cluster 7), Charolais (cluster 4), Limousine (cluster 6) and Hereford (cluster 5). The gene flow between the above breeds expressed by the fact that clusters 1, 4, 5, 6 and 7 separately represent a source of migrants but at the same time accept the flow of genes from other populations. The gene pools of Belgian Blue (cluster 2) and Blonde d'Aquitaine (cluster 3) were not affected by other breeds under consideration, and therefore they did not show four modes of gene flow. Figure 1 illustrates their contribution into the genomes of Red Angus, Angus, Charollais, Limousin and Hereford.

The Slovak populations of Charolais and Limousine achieved a relatively high proportion of genotypes that have sequence signatures from other clusters, which indicates extensive migration events between these populations. The inter-clusters gene flow ranged from 0.03% to 8.90%. The analysis of gene flow showed that only 2% of the Charolais genome was affected by other breeds. The majority of genes come from Limousine and Blonde d'Aquitaine cattle, with a contribution of 0.79% and 0.42%, respectively.

The Limousine genome showed a higher value of gene flow compared to Charolais, where up to 5% of introduced DNA were obtained from other populations. The two significant sources of gene flow come from Charolais and Blonde d'Aquitaine, with a contribution of 3.50% and 0.82%, respectively.

Belgian Blue and Blonde d'Aquitaine did not show a significant level of gene flow from the other clusters. The Angus had a higher gene flow from the Charolais (1.20%) compared to the Limousine (0.43%). Red Angus had the highest gene flow from the Angus (8.90%) as expected. On the contrary, Hereford had the main source of gene flow from Charolais (1.10%).

Our results show that the current breeding standard of the Charolais and Limousine breeds is the result of historical cooperation in the formation of these breeds. According to the results from Figure 1, the Limousine and Charolais breed did not participate in the formation of the Blonde d'Aquitaine breed, respectively, indicate that its gene pool was the only rarely influenced by other breeds. However, we know from history that the Southwestern populations of French cattle, including the Limousine, participated in the formation of the Blonde d'Aquitaine breed. Ritchie (2009) confirm our results, as there are historical branches between the Blonde d'Aquitaine and Charolais. Since can be seen from the results that the Blonde d'Aquitaine occurred in Limousine and Charolais clusters as the two major

sources of gene flow, we can say that there is an exchange between the given populations. We believe that the gene flow for the Blonde d'Aquitaine is less than 0,18%, and therefore, the program does not display it. Ouchene-Khelifi et al. (2018) confirm our opinion when they state that gene flow with a value lower than 0.01 was not displayed due to pruning to improve the readability of the images.

Figure 1. The gene flow graph in evaluated beef populations. (Angus = Cluster 1, Belgian Blue = Cluster 2, Blonde d'Aquitaine– Cluster 3, Charolais – Cluster 4, Hereford – Cluster 5, Limousine – Cluster 6 and Red Angus – Cluster 7)

Based on our results was created the maximum-likelihood (ML) phylogenetic ancestry graph expressing gene flow edges among populations. The results indicated the formation of three groups (Figures 2), but the distribution of the individual populations was slightly different compared to Bayesian analysis. Figure 2. represents the ML tree for seven evaluated beef populations, including migration arrows. Migration arrows are coloured according to the weight and the degree of admixture estimated among the evaluated breeds. The amount of gene flow between populations was expressed using a horizontal axis scale and individual horizontal branches. The standard error was stated as 10x the average standard error of the covariance matrix of a given sample. All breeds showed relatively long branches in the ML graph, except the Charolais, Belgian Blue and Blonde d'Aquitaine (Figure 2), indicating that these breeds were carried away similarly as

reported by Rochus et al. (2017). It can be seen that the Limousine and Blonde d'Aquitaine are more genetically linked and, besides, appeared on the same branch. The results point to the fact that the Limousine was involved in the formation of Blonde d'Aquitaine and have common historical ancestors. As expected, the Angus and Red Angus are more genetically linked together and appeared on the same branch as the Hereford breed similarly as reported by Barbato et al. (2020). The Charolais breed is separated from other breeds under consideration.

Figure 2 shows that the Charolais and Blonde d'Aquitaine are genetically linked, occurring on the same branch as the Limousine. After the addition of migration events, we observed the influence of Limousine on Blonde d'Aquitaine, which suggests that introgressive events have historically occurred between these breeds, which is consistent with the results of BAPS analysis. When migration events were observed, the average migration edges had a weight less than 0.2, the only exception being the edge linking the ancestors' Limousine and Blonde d'Aquitaine. This migration event reached a value higher than 0.4, indicating that the donor population contributes significantly to the recipient population as reported by Orozco-ter Wengel et al. (2015). The Charolais had a low weight of the migration edge from the Red Angus (0.2) and vice-versa. Our results most likely pointed to a historical crossbreeding to create a hybrid of these two breeds (Keane, 2011) with high carcass and performance traits (Nelson, 2015).
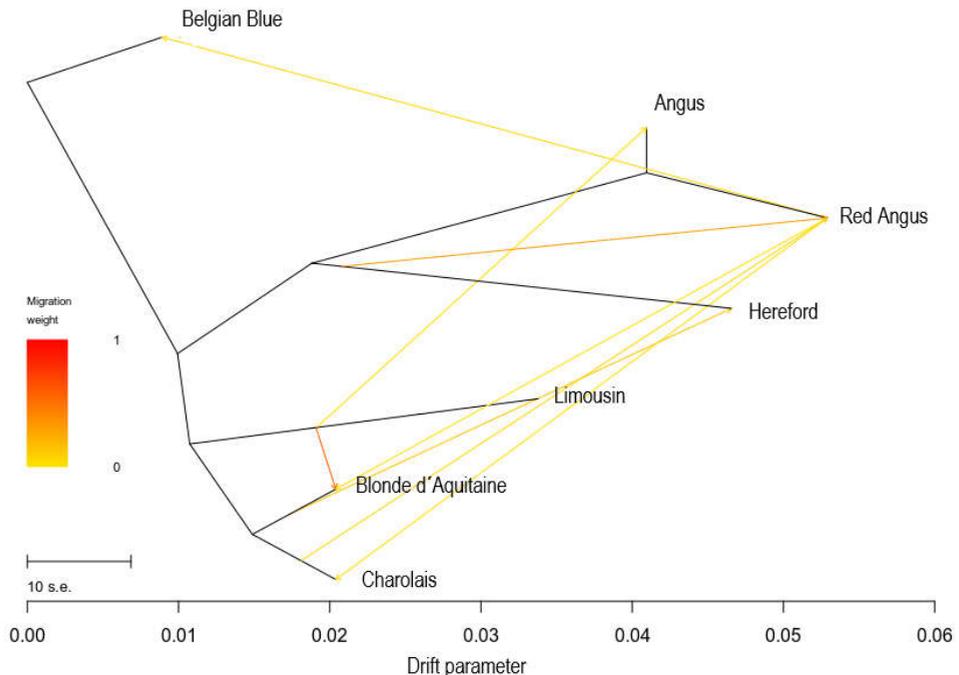
Figure 3. The maximum-likelihood (ML) tree represented the inferred relationship between analysed beef populations (with migrations settings)

CONCLUSION

The current state of modern breeds genomes is the result of lengthy and complex processes. With the help of new advanced analyses, it is possible to reveal these processes that have been involved in the formation of current cattle breeds. Obtained results indicated that the Limousine genome was influenced mainly by the Charolais genes and vice-versa. Both applied methodologies confirmed the historical contribution of Charolais cattle to the genetic make-up of Red Angus. Moreover, the Charolais genome showed genetic signatures of crossbreeding with Blonde d'Aquitaine in the 60s of the 19th century. In general, the results of this study provided knowledge about the degree of gene flow and genetic admixture among evaluated breeds, mainly due to migration events and crossbreeding during grading-up of the particular breeds. Future genome-wide association studies involving haplotypes unique for each breed can be a good basis for genomic improvement of specific carcass and performance traits.

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REFERENCES

Association Of Slovak Spotted Cattle Breeders – Cooperative (2020). History Of Breed Origin, Year 2020. Association Of Slovak Spotted Cattle Breeders – Cooperative, Retrieved Jun 20, 2020 from https://www.simmental.sk/o-plemene/historia-vzniku-plemena.html

Barbato M., Reichel M.P., Passamonti M., Low W.Y., Colli L., Tearle R., Williams J.L., Marsan P.A. (2020). A genetically unique Chinese cattle population shows evidence of common ancestry with wild species when analysed with a reduced ascertainment bias SNP panel. PLoS ONE, no. 15, vol. 4: e0231162. https://doi.org/10.1371/journal.pone.0231162

Bohórquez M.D., Ordoñez D., Suárez C.F., Vicente B., Vieira C., López-Abán J., Muro A., Ordóñez I, Patarroyo M.A. (2020). Major Histocompatibility Complex Class II (DRB3) Genetic Diversity in Spanish Morucha and Colombian Normande Cattle Compared to Taurine and Zebu Populations. Frontiers in Genetics, vol. 10, pp. 1293.

Bolnick D.I., Nosil P. (2007). Natural Selection in Populations Subject to a Migration Load. Evolution. no.61, vol. 9, pp 2229-2243. doi:10.1111/j.1558-5646.2007.00179.x.

Chang Ch.C., Chow C.C., Tellier L.C.A.M., Vattikuti S., Purcell Sh.M., Lee J.J. (2015). Second-generation PLINK: rising to the challenge of larger and richer datasets. GigaScience, vol. 4, pp.7. https://doi.org/10.1186/s13742-015-0047-8

Corander J., Tang J. (2007). Bayesian analysis of population structure based on linked molecular information. Math Biosci, vol. 205, pp. 19-31.

Goszczynski D.E., Ripoli M.V., Takeshima S.N., Baltian L., Aida Y., Giovambattista G. (2014). Haplotype determination of the upstream regulatory region and the second exon of the BoLA-DRB3 gene in Holstein cattle. Tissue Antigens vol. 83, pp. 180-183.

Groeneveld, L.F., Lenstra J. A., Eding H., Toro M. A., Scherf B., Pilling D., Negrini R., Finlay E.K., Jianlin H., Groeneveld E., Weigend S., The GLOBALDIV Consortium. (2010). Genetic diversity in farm animals – a review. Anim. Genet., vol. 41, pp. 6-31.

Hlongwane N.L., Hadebe K., Soma P., Dzomba E.F., Muchadeyi F.C. (2020). Genome Wide Assessment of Genetic Variation and Population Distinctiveness of the Pig Family in South Africa. frontiers in Genetics, vol. 11, pp. 344. doi: 10.3389/fgene.2020.00344

Kadlečík O., Pavlík I., Moravčíková N., Kasarda R. (2016). Inbreeding and genetic diversity loss of four cattle beef breeds in Slovakia. Acta fytotechn zootechn, vol. 19, pp. 59-63.

Keane M.G. (2011). Ranking of Sire Breeds and Beef Cross Breeding of Dairy and Beef Cows, Grange Beef Research Centre Occasional Series no.9, Teagasc, 2011.

Nelson P. (2015). Making Bucks From Buckskins.Charolais edge Value-added solutions. Retrieved Jun 20, 2020 https://charolaisusa.com/pdf/articles/MakingBucksFromBuckskinsFebMarch2015Edge.pdf

Kijas J.W., Townley D., Dalrymple B.P., Heaton M.P., Maddox J.F., McGrath A., Wilson P., Ingersoll R.G., McCulloch R., McWilliam S., Tang D., McEwan J., Cockett N., Oddy V.H., Nicholas F.W., Raadsma H., for the International Sheep Genomics Consortium. (2009). A genome wide survey of SNP variation reveals the genetic structure of sheep breeds. PLOS ONE, vol. 4. https://doi.org/10.1371/journal.pone.0004668

Kukučková V., Moravčíková N., Curik I., Simčič M., Mészáros G., Kasarda R. (2017). Genetic diversity of local cattle. Acta biochimica Polonica, vol. 65, pp. 421-424

Makanjuola R.O., Taylor-Robinson A.W. (2020). Improving Accuracy of Malaria Diagnosis in Underserved Rural and Remote Endemic Areas of Sub-Saharan Africa: A Call to Develop Multiplexing Rapid Diagnostic Tests. Scientifica (Cairo). 2020:3901409.

Masel J. (2011). Genetic drift. Current Biology. Cell Press., vol. 21, no. 20, pp. R837-8.

McTavish E.J., Decker J.E., Schnabel R.D., Taylor J.F., Hillis D.M. (2013). New world cattle show ancestry from multiple independent domestication events, Proc Natl Acad Sci USA, vol.110, no.15, pp.1398-1406. https://doi.org/10.1073/pnas.1303367110

Merilä J. (2014). Lakes and ponds as model systems to study parallelevolution. *J. Limnol*., vol.73, pp. 33-45.

Orozco-terWengel P., Barbato M., Nicolazzi E., Biscarini F., Milanesi M., Davies W., Williams D., Stella A., Ajmone-Marsan P., Bruford M.W. (2015). Revisiting demographic processes in cattle with genome-wide population genetic analysis. Front Genet., vol.6, pp. 191.

Ouchene-Khelifi N.A., Lafri M., Pompanon F., Ouhrouch A., Ouchene N., Blanquet V., Lenstra J.A., Benjelloun B., Da Silva A. (2018). Genetic homogeneity of North-African goats. PLoS ONE, vol. 13, no. 8, pp. e0202196. https://doi.org/10.1371/journal.pone.0202196

Pickrell J.K., Pritchard J.K. (2012). Inference of population splits and mixtures from genome-wide allele frequency data. PLoS Genet, vol. 8: e1002967.

R CORE TEAM. (2014). R: A language and environment for statistical computing. R Foundation for Statistical Computing. [Internet]. Vienna, Austria; 2013. [cited 2020 May 8] Available from: http://www.R-project.org/.

Ritchie H. (2009). Breeds of Beef and Multi-Purpose Cattle. Michigan State University. Retrieved Jun 19, 2020 from https://www.canr.msu.edu/ans/uploads/files/Breeds%20of%20Beef%20Cattle%20Ritchie%20Jan2009.pdf

Rochus C.M., Johansson A.M. (2017). Estimation of genetic diversity in Gute sheep: pedigree and microsatellite analyses of an ancient Swedish breed. Hereditas, vol. 154, pp. 4.

Tang J, Hanage W.P., Fraser Ch., Corander J. (2009). Identifying cur-rents in the gene pool for bacterial populations using an integra-tive approach. PLoS Comput Biol., vol. 5, no. 8: e1000455. http://dx.doi.org/10.1371/journal.pcbi.1000455

Upadhyay M., Bortoluzzi C., Barbato M., Marsan P.A., Colli L., Ginja C., et al. (2019). Deciphering the patterns of genetic admixture and diversity in southern European cattle using Genome-wide SNPs. Evol Appl. John Wiley & Sons, Ltd (10.1111); 2019.

Vargas J, Landi V, Martínez A, Gómez M, Camacho ME, Álvarez LÁ, Aguirre L., Delgado J.V. (2016). Molecular study of the amazonian Macabea cattle history. PLoS One,vol.11, pp.1-16.