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DEVELOPMENT AND VALIDATION OF A LOW DENSITY SNP PANEL FOR ASSESSMENT OF GENETIC DIVERSITY OF THE REINDEER (*Rangifer tarandus*) POPULATIONS

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Abstract

Reindeer (*Rangifer tarandus*) is a valuable member of the Arctic ecosystems and the main livestock species of the Russian North, which require the analysis of the genetic structure and the possibility of addressing the differences between wild and domestic forms, breeds and populations using modern molecular genetic approaches. The use of DNA chips based on parallel genotyping of hundreds of thousands of SNP markers is an effective approach to study the reindeer genome, but at the same time due to a high price, it is not beneficial for wide practical application. In this regard, the aim of our work is to select the optimal number of SNP markers that allow conducting population and genetic studies of reindeer without loss of bio-informatics content. The sample collection included wild deer (WLD, $n = 83$) inhabiting the Taimyr Peninsula and the Republic of Sakha (Yakutia), and domestic deer of the Nenets breed from the Nenets Autonomous Okrug (NEN, $n = 100$) and the Murmansk Region (MUR, $n = 19$), as well as from Even and Evenki breeds from the Republic of Sakha (Yakutia) (YAK, $n = 19$). All deer were genotyped using a high-density DNA chip BovineHD BeadChip (777,962 SNPs). After quality control and filtering, 4456 polymorphic SNP markers remained in the analysis. In the TRES program, using the Delta method, 368 of the most informative SNP markers were selected. Data processing was performed in the Admixture 1.3, PLINK 1.9 programs and R packages (ggplot2, adegenet 1.3-1, pophelper, diveRsity). It was shown that 70 % from 368 selected SNPs had a high minor allele frequency (MAF ≥ 0.3), while about 50 % from set including 4456 markers had MAF ≤ 0.1. Comparing the results of principal component analysis (PCA), discriminant principal component analysis (DAPC), and cluster analysis, no loss of information value was found for 368 SNPs compared to using the set of 4456 markers. Comparing

pairwise F_{ST} values between the studied groups of reindeer, the similarity of the interpopulation linkages was demonstrated, based on 4456 and 368 SNP markers, respectively. Thus, the selected panel of SNP markers is an informative, universal for both wild and domestic deer and a cheap approach for creating a custom DNA chip for reindeer.

Keywords: *Rangifer tarandus*, reindeer, SNP markers, DNA chips

Reindeer (*Rangifer tarandus*) is a unique species, among which both the wild and the domestic forms coexist and are valuable members of the Arctic ecosystems [1, 2]. Both the forms require the development of rational programs to conserve their genetic diversity. Rapid improvements and increased availability of new-generation sequencing technologies (next-generation sequencing) have led to an increase in the numbers of full genomes determined for various animal species each year [3, 4].

Over the past few years, numerous studies have been performed to map the genome of the reindeer, which has revealed three versions of the genome assembly. In 2017, a group of Chinese researchers introduced the first version of the domestic reindeer genome [5]. DNA was extracted from the blood of one female of the Evenk breed from a population which had been introduced into the mountains of Inner Mongolia in China, and sequencing of the sample was performed on the HiSeq 4000 platform (Illumina, San Diego, CA, USA).

This assembly showed that the total genome size of the domestic reindeer was 2.64 Gb. A total of 21,555 protein-coding genes were decoded, and 3803 genes were completely reconstructed (92.6% of the genome size). However, it was later discovered that this genome assembly was fragmented and contained a high percentage of missing data [6].

In 2019, the genome of the caribou, the North American relative of *R. tarandus* [6] with a genome size of 2.205 Gb, was announced. According to Taylor et al. [6], the caribou genome includes 33,177 protein-coding genes, including 3820 (93.1%) completely reconstructed genes. However, since the wild and the domestic forms of *R. tarandus* are clearly differentiated by single-nucleotide polymorphism (SNP) markers [8], this described genome assembly may not be relevant to the domestic reindeer. Finally, a group of Scandinavian researchers [7] performed deep sequencing of the domestic reindeer genome using the HiSeq2500 and HiSeq 4000 systems (Illumina). However, the complete nucleotide sequence of this assembly, which had a total size of 2.66 Gb and included 26,785 protein-coding genes, cannot be found in the National Center for Biotechnology Information database and other platforms. As a result, despite significant successes in decoding the reindeer genome, this species remains in the non-model category.

Negative anthropogenic factors and unpredictable climate changes in the reindeer habitat have led to irreversible critical changes in the biodiversity of this species [9, 10]. At present, there has been an active research for the search and implementation of various approaches for assessment of the genetic diversity, for the study of the population structure, and for the reliable differentiation of the intraspecific groups [11]. SNPs have been widely used as DNA markers for studying the genetic structure of populations [12]. The use of a DNA microarray enables simultaneous genotyping of myriad markers. However, the main task in most studies is to determine the pedigree, the group, or the individual origin of the animal [13]. In this case, it is reasonable to use only those SNPs that can most accurately differentiate relevant individuals at a lower cost using faster procedures compared to that in genome-wide genotyping approaches [13, 14]. In this study, SNPs that met the criteria as markers for creating a custom DNA microarray were selected for the reindeer using a minor allele frequency (MAF) above 0.3 to preserve the bioinformatics value and universal nature of this infor-

mation in population genetics studies of both the domestic and the wild forms of *R. tarandus*.

Our aim was to select the optimal number of SNPs that enabled population genetics studies of the reindeer without the loss of bioinformatics content and for performing a comparative analysis of the information of the selected and of all the polymorphic SNPs.

Techniques. The studies were performed on the wild and the domestic reindeer in 2019. The wild reindeer population (WLD, $n = 83$) included the reindeer inhabiting the western part of the Taimyr Peninsula and representatives of the Leno-Olenek and the Sundrun subpopulations from the Republic of Sakha (Yakutia) territory.

The domestic reindeer group consisted of animals of the Nenets breed, which are bred in the territory of the Nenets Autonomous district (NEN, $n = 100$ and the Murmansk Region (MUR, $n = 19$), as well as the Even and the Evenk breeds from the Republic of Sakha (Yakutia) (YAK, $n = 19$). Genomic DNA was extracted using the Extran2 DNA kits (Syntol CJSC, Moscow, Russia) according to the manufacturer's instructions. The quality and integrity of the DNA samples were checked by electrophoresis on a 1% agarose gel. DNA concentration was measured on a Qubit 2.0 fluorimeter (Invitrogen, Carlsbad, CA, USA). DNA purity was determined by evaluating the absorption ratio of A_{260}/A_{280} on a NanoDrop8000 spectrophotometer (Thermo Fisher Scientific, Waltham, MA, USA). All the investigated reindeer individuals were genotyped using the Illumina BovineHD BeadChip, which contained 777,962 SNPs (Illumina). Genotypes were obtained using the Genome Studio 2 (Illumina). Reading quality (GenCall, GC) and the degree of SNP clustering (GenTrain, GT) were evaluated by establishing filters with limit values of 0.5 for both indicators [15]. In PLINK v. 1.90 [16], SNPs located on the sex chromosomes, characterized by MAF below 5% (-MAF 0.05), that deviated from the Hardy-Weinberg equilibrium at $p < 10^{-6}$ (-hwe $1e^{-6}$), those that were in linkage disequilibrium (--indep-pairwise 50 5 0.5), and those for which less than 90% of the animals were genotyped (- -geno0.1) were excluded from the analysis. After filtering, 4456 polymorphic SNPs were included for analysis.

The next step was selection of the most informative SNPs for inclusion in the user panel. Markers were selected in the TRES program (Toolbox for Ranking and Evaluation of SNPs) [13] according to the Delta method [17] in accordance with the equation:

$$\delta = |p^i_A - p^j_A|,$$

where p^i_A is the frequency of allele A in the i^{th} population and p^j_A is the frequency of the same allele in the j^{th} population.

Next, the loci were evaluated based on their δ values and ranked in a descending order from the highest values to the lowest value of 0.2. As a result, 368 SNPs were selected.

The graphs of the distribution of SNPs in the different groups depending on the MAF values were visualized using the R package [18]. Principal component analysis was performed in PLINK 1.9 and the results were visualized in the R package ggplot2 [19]. Discriminant analysis of the principal components for the investigated reindeer groups was performed using the R package adegenet 1.3-1 [20, 21], the cluster analysis — in Admixture 1.3 [22] with subsequent visualization with R package «pophelper» [23]. The pairwise fixation index (F_{ST}) values [24] were calculated with R package diveRsity [25]. All bioinformatics analyses were performed for the 4456 SNPs and 368 SNPs to evaluate the effectiveness of the proposed panel (368 SNPs) compared to the complete set of polymorphic SNPs (4456) identified using the Illumina BovineHD BeadChip for

the reindeer populations.

Results. The distribution of the selected SNPs among the groups were compared depending on the MAF values (Fig. 1), which revealed that the group with MAF of 0.1 contained the largest number of SNPs in the panel of 4456 markers (approximately 49.3%), whereas the number of markers in the remaining groups decreased from 800 in the group with MAF of 0.2 (17.9%) to 250 (5.6%) in the group with MAF of 0.5 (see Fig. 1, A). When using a panel of 368 markers, the distribution of SNPs was drastically different (see Fig. 1, B). Thus, the group with MAF of 0.1 accounted for only 4.1% of the total number of panel markers. The largest number of SNPs (57.1%) was represented by the groups with MAF values of 0.3 and 0.4. The group with MAF of 0.5 comprised approximately 15.8% of all panel markers.

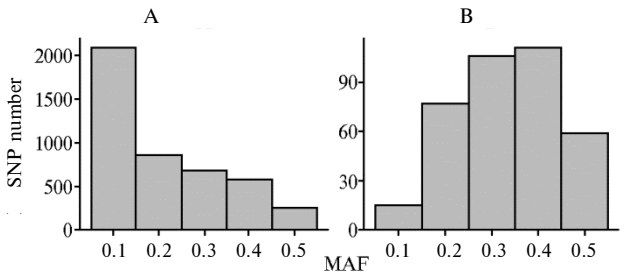

Fig. 1. Distribution of the SNPs by group (2019) **depending on the minor allele** (MAF) **frequency by comparison of two marker panels:** A — final data set comprised 4456 SNPs for the domestic and the wild reindeer (*Rangifer tarandus*) from the BovineHD BeadChip; B — 368 SNPs selected to create a custom DNA microarray.

In general, principal component analysis of the investigated reindeer groups based on 4456 SNPs and 368 SNPs revealed no significant differences in the character of the spatial formation of clusters (Fig. 2, A, B). Thus, the first main component (PC1) clearly separated the MUR and the NEN groups from the WLD group, whereas the YAK population, which was nearly located on PC1, was differentiated by the second main component (PC2) among the three reindeer clusters. However, the genetic variability magnitude explained by PC1 and PC2 when using a panel of 4465 SNPs (see Fig. 2, A) was several-fold lower compared to that in the second panel (see Fig. 2, B): 11.8% vs. 36.28% for PC1 and 3.21% vs. 10.32% for PC2.

The discriminant analysis of the principal components was used to examine the patterns in the genetic diversity of the investigated reindeer groups (Fig. 2, C, D). Linear discriminant 1 (LD 1) clearly differentiated the NEN, the MUR, and the YAK populations from the WLD group. The YAK group was the most distant for linear discriminant 2 (LD 2). In addition, the NEN, the MUR, and the WLD groups were close to the LD 2 axis. No significant differences were found in the information of the two investigated systems.

Comparison of the degree of genetic differences between the investigated reindeer groups (Table) showed the similarity of interpopulation relationships estimated using the 4456 SNPs and 368 SNPs. Thus, the highest genetic closeness was observed between the MUR and the NEN groups, and the maximum differentiation was found between the WLD and the NEN.

The results of the ADMIXTURE-analysis for the investigated reindeer groups based on the 4456 SNPs (see Fig. 3, A) and 368 SNPs (see Fig. 3, B) for the most probable number of clusters 2 and 3 have been shown in Figure 3. At K = 2, both panels showed similar clustering results. The NEN and the WLD populations were clearly differentiated and formed their own cluster (light grey for the WLD and black for the NEN), whereas the MUR and the YAK populations represented mixed clusters. At K = 3, the YAK group segregated into its own cluster, and presented a consolidation of individuals within which looked more identically by using a kit including all polymorphic SNPs (see Fig. 3, A).

However, differences in the pattern of distribution of the animal groups at K = 3 were insignificant.

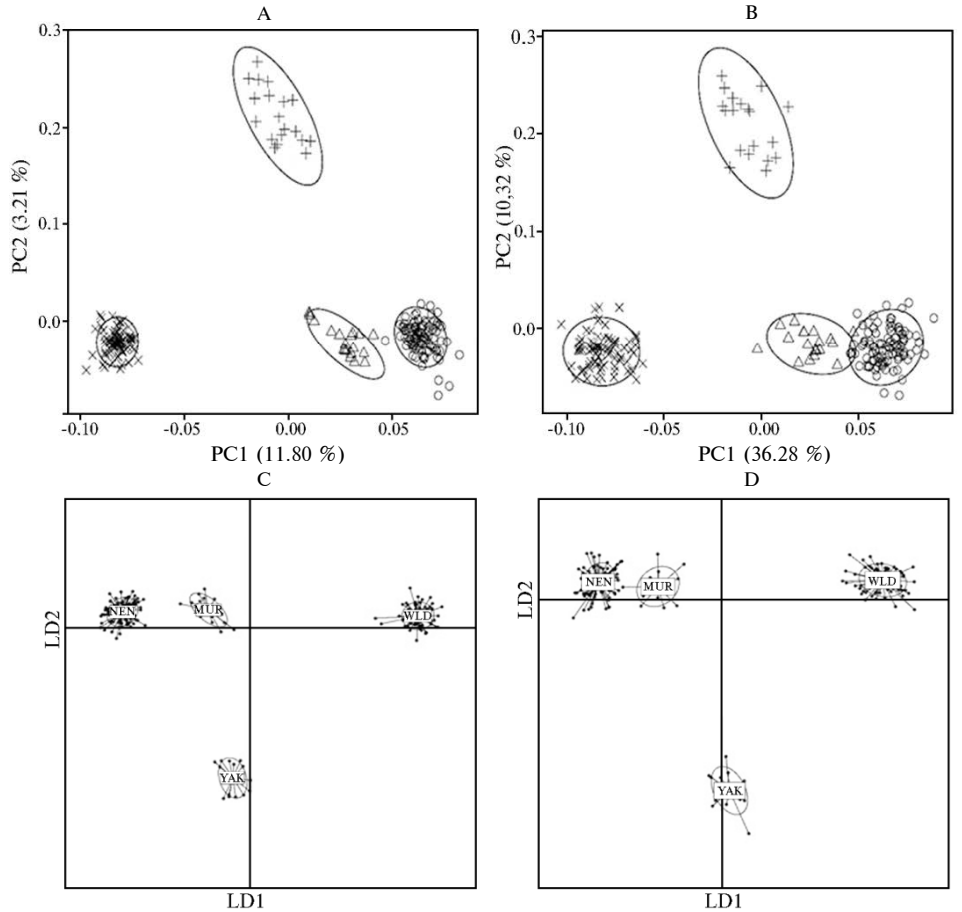

Fig. 2. Results of the principal component analysis (A, B) **and discriminant analysis of the principal components** (C, D)**, conducted for the four populations of reindeer (*Rangifer tarandus*) based on data from two SNP panels:** A, C — final data set comprised of 4456 SNPs detected with the BovineHD BeadChip; B, D — 368 SNPs selected to create a custom DNA microarray; WLD — wild population (the Taimyr Peninsula, the Republic of Sakha-Yakutia), NEN — domestic reindeer of the Nenets breed (the Nenets Autonomous Okrug), MUR — domestic reindeer of the Nenets breed (the Murmansk region), YAK — domestic reindeer of the Even and Evenk breeds (the Republic of Sakha-Yakutia) (2019).

The effectiveness of using a DNA microarray designed for cattle to evaluate the reindeer has been demonstrated in foreign [26, 27] and domestic studies [8, 28]. Nevertheless, among all the genotyped SNPs, only approximately 5% of the SNPs are polymorphic and have been directly used for analysis: 1068 for the black-tailed and the white-tailed deer [26], 1532 for the representatives of the genus Cervus [27], and 512 for the Yakut domestic reindeer [28] when using the Bovine SNP50 BeadChip and 8357 and 8145, respectively, for the domestic and the wild reindeer from among 777,962 SNPs on the BovineHD BeadChip [8].

Despite the success of using these DNA microarrays to characterise the genetic structure of the reindeer populations and assess their genetic diversity and relationships, there are several factors that prevent the use of this approach for routine testing. First, the DNA microarray is costly. Second, nearly 95% of the content of a DNA microarray is uninformative for the reindeer. In addition, detailed genomic studies are not always required for practical applications in the reindeer husbandry.

F_{ST} values calculated from 4456 and 368 SNPs in the four reindeer populations (*Rangifer tarandus*) (2019)

Population	NEN	MUR	YAK	WLD
NEN		0.025	0.184	0.218
MUR	0.012		0.142	0.138
YAK	0.043	0.040		0.215
WLD	0.060	0.042	0.048	

N o t e. WLD — wild population (the Taimyr Peninsula, the Republic of Sakha-Yakutia), NEN — domestic reindeer of the Nenets breed (the Nenets Autonomous Okrug), MUR — domestic reindeer of the Nenets breed (the Murmansk region), YAK — domestic reindeer of the Even and Evenk breeds (the Republic of Sakha-Yakutia); F_{ST} — fixation index. The values calculated for 368 SNPs are above the diagonal, the values calculated for the 4456 SNPs are below the diagonal.

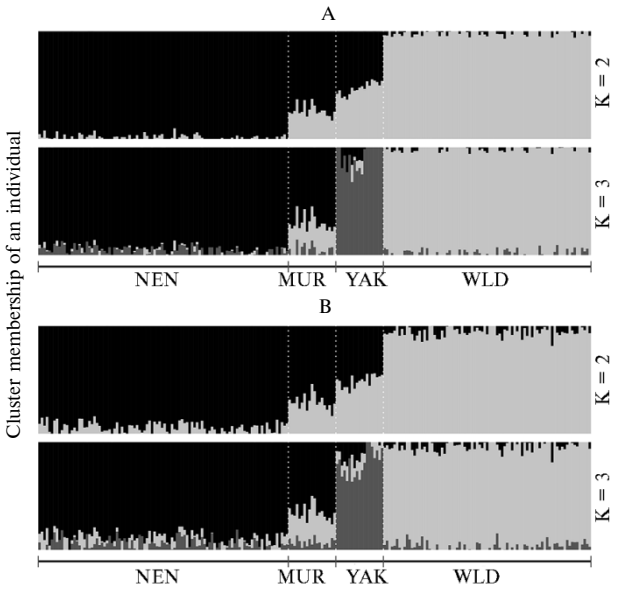

Fig. 3. Comparative ADMIXTURE analysis of the four reindeer populations (*Rangifer tarandus*) (2019) **for two SNP panels:** A — final data set comprised 4456 SNPs detected with the BovineHD BeadChip; B — 368 SNPs selected to create a custom DNA microarray; K — the number of most probable clusters; WLD — wild population (the Taimyr Peninsula, the Republic of Sakha-Yakutia), NEN — domestic reindeer of the Nenets breed (the Nenets Autonomous Okrug), MUR — domestic reindeer of the Nenets breed (the Murmansk region), YAK — domestic reindeer of the Even and the Evenk breeds (the Republic of Sakha-Yakutia).

In this case, the development of a custom DNA microarray may reduce the cost of analysis without significantly decreasing its biological significance [29, 30]. Similar SNP panels containing a limited number of informative markers have been designed for model farm animals, such as the sheep [31, 32], cattle [33], and horses [34].

Our study showed that 368 SNPs provided reproducible information for all detected polymorphic markers (4456), which was found to be consistent with previously obtained data for other mammalian species. According to Heaton et al. [33], 32 SNPs are sufficient to determine the origin with an accuracy of 99.99% in the beef cattle populations (even in cases of highly mixed herds). Kijas et al. [31] suggested using SNP panels of 88 markers for paternity confirmation in more than 80 sheep breeds, and using 101 markers in horse breeding for the same purposes [34]. In addition, a panel of 163 markers is sufficient for determining the breed and individual affiliation of a wide range of sheep breeds [32].

The MAF is an important parameter in selecting SNPs for the subsequent development of an informative custom DNA microarray. Using the TRES bioinformatics approach [13], we selected 368 SNPs from among 4456 polymorphic SNPs. The calculations showed bias in the selected SNPs towards increasing MAFs from 0.1 to 0.3-0.4. The observed trend was consistent with that obtained by Kijas et al. [31] and Heaton et al. [32] in the development of panels for determining the origin of sheep (panels consisted of 88 and 163 markers, respectively). According to Kijas et al. [31], choosing SNPs with a shift towards an MAF $\geq$ 0.3 for inclusion in the panel enhances the versatility of the panel, ena-

bling its application to diverse breeds and population compositions of the investigated species. The 368 markers selected in this study met this criterion.

We selected SNP markers from a DNA microarray developed for the cattle, in accordance with their localisation along the chromosomal characteristics of the cattle. As the alignment of the nucleotide sequences of the reindeer genome was performed using the cattle genome as a reference [6] (without breakdown into the reindeer chromosomes), the SNP panel can provide the same amount of information as those developed based on the reindeer genome assembly proposed by Taylor et al. [6].

Thus, the minimum required number of SNPs was selected, allowing for genetic studies in the reindeer populations without the loss of bioinformatics content. To assess the information of a panel of 368 SNPs, the data obtained were compared to the results calculated using 4456 SNPs identified by a high-density microarray. Among the markers included in the test panel, a clear bias towards SNPs with MAF of more than 0.3 was observed, corresponding to international criteria for creating panels for various types of agricultural animals. All bioinformatics approaches (the principal component analysis, the discriminant analysis of principal components, the cluster analysis, and calculation of pairwise genetic distances by F_{ST} values) showed no loss in efficiency of the 368 SNPs panel. Successful testing of the presented panel was performed in the three breeds of the domestic reindeer (the Nenets, the Even, the Evenk) and in the two populations of the wild reindeer (the Taimyr and the Yakut), demonstrating the universal nature of the selected SNPs. Selected SNPs may be recommended for inclusion during construction of a custom DNA microarray designed for genomic characterisation of the wild and the domestic reindeer.

REFERENCES

1. Baskin L.M. *Severnyi olen'. Upravlenie povedeniem i populyatsiyami. Olenevodstvo. Okhota* [Reindeer. Manage behavior and populations. Reindeer husbandry. Hunting]. Moscow, 2009 [in Russ.).
2. Laishev K.A., Yuzhakov A.A. *Nauchnyi vestnik Yamalo-Nenetskogo avtonomnogo okruga*, 2017, 1[94): 45-48 [in Russ.).
3. Zhang J., Chiodini R., Badr A., Zhang G. The impact of next-generation sequencing on genomics. *Journal of Genetics and Genomics,* 2011, 38[3): 95-109 [doi: 10.1016/j.jgg.2011.02.003).
4. Kchouk M., Gibrat J.F., Elloumi M. Generations of sequencing technologies: from first to next generation. *Biol. Med. [Aligarh)*, 2017, 9[3): 1000395 [doi: 10.4172/0974-8369.1000395).
5. Li Z., Lin Z., Ba H., Chen L., Yang Y., Wang K., Qiu Q., Wang W., Li G. Draft genome of the reindeer [*Rangifer tarandus*). *GigaScience*, 2017, 6[12): 1-5 [doi: 10.1093/gigascience/gix102).
6. Taylor R.S., Horn R.L., Zhang X., Golding G.B., Manseau M., Wilson P.J. The caribou [*Rangifer tarandus*) genome. *Genes*, 2019, 10[7): 540 [doi: 10.3390/genes10070540).
7. Weldenegodguad M., Pokharel K., Ming Y., Honkatukia M., Peippo J., Reilas T, Røed K.H., Kantanen J. OP149 sequencing of reindeer [*Rangifer tarandus*) genomes: insights into evolution, domestication, and adaptation. *Proc. 37th International Conference on Animal Genetics.* Lleida, Spain, 2019: 42.
8. Kharzinova V.R., Dotsev A.V., Deniskova T.E., Solovieva A.D., Fedorov V.I., Layshev K.A., Romanenko T.M., Okhlopkov I.M., Wimmers K., Reyer H., Brem G., Zinovieva N.A. Genetic diversity and population structure of domestic and wild reindeer [*Rangifer tarandus* L. 1758): a novel approach using BovineHD BeadChip. *PLoS ONE*, 2018, 13[11): e0207944 [doi: 10.1371/journal.pone.0207944).
9. Yannic G., Pellissier L., Ortego J., Lecomte N., Couturier S., Cuyler C., Dussault C., Hundertmark K.J., Irvine R.J., Jenkins D.A., Kolpashikov L., Mager K., Musiani M., Parker K.L., Røed K.H., Sipko T., Þórisson S.G., Weckworth B.V., Guisan A., Bernatchez L., Côté S.D. Genetic diversity in caribou linked to past and future climate change. *Nature Climate Change*, 2014, 4: 132-137 [doi: 10.1038/nclimate2074).
10. Yuzhakov A.A. *Materialy V Mezhdunarodnoi nauchnoi konferentsii «Ekologiya drevnikh i traditsionnykh obshchestv»* [Proc. V Int. Conf. «Ecology of ancient and traditional societies»]. Tyumen', 2016: 269-274 [in Russ.).
11. Ogden R. Unlocking the potential of genomic technologies for wildlife forensics. *Molecular Ecology Resources*, 2011, 11[Suppl. 1): 109-116 [doi: 10.1111/j.1755-0998.2010.02954.x).
12. Helyar S.J., Hemmer-Hansen J., Bekkevold D. Taylor M.I., Ogden R., Limborg M.T., Car-

iani A., Maes G.E., Diopere E., Carvalho G.R., Nielsen E.E. Application of SNPs for population genetics of nonmodel organisms: new opportunities and challenges. *Molecular Ecology Resources*. 2011, 11[Suppl. 1): 123-136 [doi: 10.1111/j.1755-0998.2010.02943.x).
13. Kavakiotis I., Triantafyllidis A., Ntelidou D., Alexandri P., Megens H.J., Crooijmans R.P.M.A., Groenen M.A.M., Tsoumakas G., Vlahavas I. TRES: identification of discriminatory and informative SNPs from population genomic data. *Journal of Heredity*, 2015, 106[5): 672-676 [doi: 10.1093/jhered/esv044).
14. Wilkinson S., Wiener P., Archibald A.L., Law A., Schnabel R.D., McKay S.D., Taylor J.F., Ogden R. Evaluation of approaches for identifying population informative markers from high density SNP chips. *BMC Genetics*, 2011, 12: 45 [doi: 10.1186/1471-2156-12-45).
15. Fan J.B., Oliphant A., Shen R., Kermani B.G., Garcia F., Gunderson K.L., Hansen M., Steemers F., Butler S.L., Deloukas P., Galver L., Hunt S., Mcbride C., Bibikova M., Rubano T., Chen J., Wickham E., Doucet D., Chang W., Campbell D., Zhang B., Kruglyak S., Bentley D., Haas J., Rigault P., Zhou L., Stuelpnagel J., Chee M.S. Highly parallel SNP genotyping. *Cold Spring Harb. Symp. Quant. Biol.*, 2003, 68: 69-78 [doi: 10.1101/sqb.2003.68.69).
16. Chang C.C., Chow C.C., Tellier L.C., Vattikuti S., Purcell S.M., Lee J.J. Second-generation PLINK: rising to the challenge of larger and richer datasets. *GigaScience*, 2015, 4[1): s13742−015−0047−8 [doi: 10.1186/s13742-015-0047-8).
17. Shriver M.D., Smith M.W., Jin L., Marcini A., Akey J.M., Deka R., Ferrell R.E. Ethnic-affiliation estimation by use of population-specific DNA markers. *American Journal of Human Genetics*, 1997, 60[4): 957-964.
18. R Core Team [2018). R: A language and environment for statistical computing. R Foundation for Statistical Computing, Vienna, Austria. Available: https://www.R-project.org/. No date.
19. Wickham H. *ggplot2 — Elegant graphics for data analysis*. Springer-Verlag, NY, 2009 [doi: 10.1007/978-0-387-98141-3).
20. Jombart T. adegenet: a R package for the multivariate analysis of genetic markers. *Bioinformatics*, 2008, 24[11): 1403-1405 [doi: 10.1093/bioinformatics/btn129).
21. Jombart T., Ahmed I. adegenet 1.3-1: new tools for the analysis of genome-wide SNP data. *Bioinformatics*, 2011, 27[21): 3070-3071 [doi: 10.1093/bioinformatics/btr521).
22. Alexander D.H., Novembre J., Lange K. Fast model-based estimation of ancestry in unrelated individuals. *Genome Research*, 2009, 19[9): 1655-1664 [doi: 10.1101/gr.094052.109).
23. Francis R.M. POPHELPER: an R package and web app to analyse and visualize population structure. *Molecular Ecology Resources*, 2017, 17[1): 27-32 [doi: 10.1111/1755-0998.12509).
24. Weir B.S., Cockerham C.C. Estimating F-statistics for the analysis of population structure. *Evolution*, 1984, 38[6): 1358-1370 [doi: 10.2307/2408641).
25. Keenan K., McGinnity P., Cross T.F., Crozier W.W., Prodöhl P.A. diveRsity: an R package for the estimation of population genetics parameters and their associated errors. *Methods in Ecology and Evolution*, 2013, 4[8): 782-788 [doi: 10.1111/2041-210X.12067).
26. Haynes G.D., Latch E.K. Identification of novel single nucleotide polymorphisms [SNPs) in deer [*Odocoileus* spp.) using the BovineSNP50 BeadChip. *PLoS ONE*, 2012, 7[5): e36536 [doi: 10.1371/journal.pone.0036536).
27. Kasarda R., Moravčíková N., Trakovická A., Krupová Z., Ondrej K. Genomic variation across cervid species in respect to the estimation of red deer diversity. *Acta Veterinaria-Beograd*, 2017, 67[1): 43-56 [doi: 10.1515/acve-2017-0005).
28. Kharzinova V.R., Dotsev A.V., Solov'eva A.D., Fedorov V.I., Okhlopkov I.M., Vimmers K., Reier Kh., Brem G., Zinov'eva N.A. Population-genetic characteristics of domestic reindeer of yakutia based on whole-genome SNP analysis. *Agricultural Biology* [*Sel'skokhozyaistvennaya Biologiya*], 2017, 52[4): 669-678 [doi: 10.15389/agrobiology.2017.4.669eng).
29. Lorenz M.G.O., Cortes L.M., Lorenz J.J., Liu E.T. Strategy for the design of custom cDNA microarrays. *Biotechniques,* 2003, 34[6): 1264-1270 [doi: 10.2144/03346mt02).
30. Neerincx P.B.T., Rauwerda H., Nie H., Groenen M.A.M., Breit T.M., Leunissen J.A.M. OligoRAP — an Oligo Re-Annotation Pipeline to improve annotation and estimate target specificity. *BMC Proc.*, 2009, 3[Suppl. 4): S4 [doi: 10.1186/1753-6561-3-S4-S4).
31. Kijas J.W., McEwan J., Clarke S., Henry H., Maddox J., McCulloch R., Driver F., Ilic K., Heaton M. *Development of a SNP panel for parentage assignment in sheep*. International Sheep Genomics Consortium, 2012. Available: http://sheephapmap.org/news/PAG_2012_ParentagePoster.pdf. No date.
32. Heaton M.P., Leymaster K.A., Kalbfleisch T.S., Kijas J.W., Clarke S.M., McEwan J., Maddox J.F., Basnayake V., Petrik D.T., Simpson B., Smith T.P., Chitko-McKown C.G., International Sheep Genomics Consortium. SNPs for parentage testing and traceability in globally diverse breeds of sheep. *PLoS ONE*, 2014, 9[4): e94851 [doi: 10.1371/journal.pone.0094851).
33. Heaton M.P., Harhay G.P., Bennett G.L., Stone R.T., Grosse WM, Casas E., Keele J.W., Smith T.P.L., Chitko-McKown C.G., Laegreid W.W. Selection and use of SNP markers for animal identification and paternity analysis in U.S. beef cattle. *Mammalian Genome*, 2002, 13[5): 272-281 [doi: 10.1007/s00335-001-2146-3).
34. Holl H.M., Vanhnasy J., Everts R.E., Hoefs-Martin K., Cook D., Brooks S.A., Carpenter M.L., Bustamante C.D., Lafayette C. Single nucleotide polymorphisms for DNA typing in the domestic horse. *Animal Genetics*, 2017, 48[6): 669-676 [doi: 10.1111/age.12608).