



Original Article

Genetic Diversity of Cattle Bred in Territory of the Tyumen Region, Russia

Kabitskaya, Ya. A¹ *, Boyko, E. G¹

1. Federal State Budget Institution of Higher Education Northern Trans-Ural State Agricultural University, Centre for Genomic Technologies, Tyumen, Russia

Received 15 July 2021; Accepted 31 July 2021
Corresponding Author: yanakabickaya@yandex.ru

Abstract

Improving the genetic potential of animals using genomic technologies is an effective way to solve problems in domestic breeding of dairy breeds. The aim of the current study is to analyze the genetic diversity of Holstein cattle bred in the Tyumen Region using microsatellite markers. The blood samples of 241 Holstein cow were collected for genomic analysis. Genetic identification was performed using COrDIS kits. The resulting data indicated the presence of genetic diversity in the studied sample of dairy cattle bred at the Tyumen Region for a number of indicators. The number of alleles at 15 microsatellite loci in the tested breeds varied from 5 to 14. Eighteen unique allelic variants can be breed-specific markers for genetic identification of dairy cattle. The recorded data showed a high level of animal genetic variability. A high level of polymorphism ($A_e \geq 3.32$) was detected in 47% of the loci under research. High values of the observed heterozygosity up to 0.91 at the Eth3 locus and of the polymorphism up to 0.77 at the Tgla53 locus were detected. In the results, 7% erroneous entries in the progeny pedigree was detected. The results of the current study contribute to the knowledge of genetic diversity of dairy cattle in the Tyumen region, Russia, and also paved the way to seek new relationships between allelic variants of microsatellite loci with the productive traits of animals.

Keywords: cattle, Holstein breed, microsatellites, genetic variety, genetic identification

Diversité Génétique des Bovins Elevés sur le Territoire de la Région de Tioumen, en Russie

Résumé: L'amélioration du potentiel génétique des animaux à l'aide des technologies génomiques est un moyen efficace de résoudre les problèmes de l'élevage domestique des races laitières. L'objectif de la présente étude est d'analyser la diversité génétique des bovins Holstein élevés dans la région de Tioumen à l'aide de marqueurs microsatellites. Les échantillons de sang de 241 vaches Holstein ont été collectés pour analyse génomique. L'identification génétique a été réalisée à l'aide de kits COrDIS. Les données résultantes ont indiqué la présence d'une diversité génétique dans l'échantillon étudié de bovins laitiers élevés dans la région de Tioumen pour un certain nombre d'indicateurs. Le nombre d'allèles à 15 loci microsatellites dans les races testées variait de 5 à 14. Dix-huit variantes alléliques uniques peuvent être des marqueurs spécifiques à la race pour l'identification génétique des bovins laitiers. Les données enregistrées ont montré un niveau élevé de variabilité génétique animale. Un niveau élevé de polymorphisme ($A_e \geq 3.32$) a été détecté dans 47% des loci étudiés. Des valeurs élevées de l'hétérozygotie observée jusqu'à 0,91 au locus Eth3 et du polymorphisme jusqu'à 0,77 au locus Tgla53 ont été détectées. Dans les résultats, 7% d'entrées erronées dans la généalogie de la descendance ont été détectés. Les résultats de la présente étude contribuent à la connaissance de la diversité génétique des bovins laitiers dans la région de Tioumen, en Russie, et ont également ouvert la voie à la recherche de nouvelles relations entre les variantes alléliques des loci microsatellites avec les traits productifs des animaux.

Mots-clés: Bovin, Race Holstein, Microsatellites, Variété Génétique, Identification Génétique

1. Introduction

Improving the genetic potential of some animal breeds using developments in domestic and adapted foreign breeds based on genetic technologies (including genomic) is an effective way to solve problems in the domestic breeding of pedigree animals. At present, in a number of European countries, obtaining reliable results of animal genotyping in several generations is a prerequisite and a necessary criterion for their use in genomic selection.

The genetic structure of cattle bred in livestock enterprises in the Tyumen region, Russia, remains poorly understood. The system of screening/selecting animals by phenotype using the results of immunogenetic analyses in small groups of animals is still used. At the same time, due to the high level of polymorphism (informativeness), the uniform distribution throughout the animal genome, and codominant inheritance, microsatellite (STR) markers are a convenient tool for studying genetic diversity, determining kinship, and identifying herds (1-3).

To obtain reliable data compatible with international identification systems on the origin of descendants according to STR markers, it is necessary and expedient to introduce DNA technologies into high-tech dairy enterprises in the region, where imported semen and animals are purchased. The data obtained on the genome of each animal will allow specialists to identify errors in the primary registration, carry out competent breeding work, and use in the future the results obtained in genomic breeding of cattle on a national scale.

The aim of the current study was to analyze the genetic diversity of Holstein cattle bred in the Tyumen region, Russia, using microsatellite markers. To achieve this goal the following tasks were identified:

- 1) Determine the genetic structure of dairy cattle herd in the Tyumen region, Russia, using microsatellite loci.
- 2) Reveal the genetic variability level of dairy cattle bred at the enterprise of the Tyumen region using microsatellite loci.

Reveal errors of primary accounting and experimental data related to the quality of the DNA test being used for genetic identification by microsatellite regions of the genome of cattle.

2. Material and Methods

The research was carried out at the Centre for Genomic Technologies of the Federal State Budgetary Educational Institution of Higher Education at the Northern Trans-Urals State Agrarian University during 2020. The biological materials for the study were blood samples from 241 head of Holstein cattle bred at a large industrial complex in the Tyumen region of Russia. Genetic identification was carried out using commercial kits of OOO Gordis (Russia), which contained a panel with microsatellite regions of the nuclear genome of animals at 15 loci: Eth3, Bm1818, Crsm60, Inra23, Tgla227, Tgla126, Tgla122, Sps115, Eth225, Tgla53, Bm224113, Bm224113, Eth10, Csm66, and Ilst6 (<https://gordiz.ru/>).

Genomic DNA was extracted from liquid and dried blood using the ion-exchange resin "Chelex 100" (Bio-Rad, USA), phenol (Sambrook), and a commercial kit of reagents, "DNA Extran" (OOO "Syntol", Russia) (4). DNA quality was checked electrophoretically using 2% agarose gel. The DNA concentration was measured using a Qubit 3.0 fluorometer (Thermo Fisher, USA), a dsDNA BR kit (Thermo Fisher, USA), and the instrument software. Samples with a DNA concentration of at least 20-40 ng/ μ L were suitable for STR analysis. PCR was

performed using a ProFlex 96-well PCR system (Applied Biosystems, Singapore). Amplification products were separated according to the manufacturer's protocol using an ABI 3500 genetic analyzer (Thermo Fisher Scientific, Japan).

The following indicators were used to assess the genetic structure: allele frequency (p); polymorphism level (A_e); observed (H_o) and expected (H_e) heterozygosities; Wright Fixation Index (F_{is}); and the information polymorphism amount (PIC) (2, 5-7).

The calculation of indicators for each locus was made by the variation statistics method using the Microsoft Office Excel 2010 software package.

3. Results and Discussion

The characteristics of 15 microsatellite loci subjected for investigation of dairy cattle animals (Holstein, Holsteinized black-and-white breeds) bred at large industrial enterprises in the Tyumen and Sverdlovsk regions are presented in Table 1.

Table 1. Characterization of microsatellite loci existing in the genome of dairy cattle

Chromosome	Locus	Reference interval of sizes of microsatellite loci fragments and nucleotide pairs (n.p.)		Range of allelic variants of microsatellite loci and nucleotide pairs (n.p.) in dairy cattle	
		theoretical	actual	Holstein breed bred at the enterprise of the Tyumen region	Holstein black-and-white breed bred at the enterprises of the Sverdlovsk region *
D2S26	Bm2113	133 – 165	125 – 143		125 – 139
D3S10	Inra23	197 – 229	198 – 220	198 – 218	198-216
D5S3	Eth10	207 – 235	209 – 225		209 – 225
D7S8	Ilst6	273 – 320	286 – 302	288 – 296	-
D9S2	Eth225	138 – 167	140 – 160		140 – 152
D1S34	Bm1824	175–202	178 – 190		178 – 190
D10S5	Csrn60	80 – 115	90 – 116	92 – 102	-
D15	Sps115		246–262	240 – 260	248-260
D16S3	Tgla53	168 – 210	154 – 186		154 – 188
D14S31	Cssm66	140 – 179	181 – 209	177 – 199	-
D18S1	Tgla227	70 – 110	77 – 103	81 – 103	77-103
D19S2	Eth3	95 – 129	109 – 131	117 – 129	109-129
D20S1	Tgla126	113 – 136	113 – 123	115 – 123	107-123
D21S6	Tgla122	137 – 190	139 – 185	141 – 183	139-183
D23S21	Bm1818	250 – 270	256 – 272	258 – 270	258-272
Total	15				

*-Literature data (3)

To characterize polymorphism, the reference interval of fragments was analyzed, and the range of allelic variants for STR markers was determined in 241 Holstein animals bred in the Tyumen region. The actual (recommended by the manufacturer of the test systems) reference range of sizes determined for fragments of nucleotide sequences belonging to the microsatellite

loci of the cattle genome slightly differed from the theoretical one (according to international identification systems). The difference in base pairs ranged from 1 to 41. The range of allelic variants for 15 studied animal loci in the breed under research ranged from 81 to 296 base pairs. The range of allelic variants in animals in the sample under investigation corresponded to the

actual reference range of fragment sizes at all microsatellite loci, except for the Tgla53 locus, the interval of which was 2 base pairs longer. Thus, for the correct genetic identification of cattle bred at the enterprise in the Tyumen region using this set of reagents, it was necessary to include an additional interval in the binary grid at the above-indicated locus.

When comparing the data obtained in this research with that available in the literature for the Holsteinized black-and-white breed (Sverdlovsk region), the coincidence of the actual reference interval for 5 loci under research (Bm2113, Eth10, Eth225, Bm1824, and Tgla53) was found. Differences in the ranges of allelic variants at loci Inra23, Tgla122, and Bm1818 by 2 base

pairs were found. Loci distinguished by 8 base pairs were Sps115, Eth3, and Tgla126 (3).

The data obtained in the current study indicated the presence of genetic diversity in the studied sample of dairy cattle bred at the Tyumen Region enterprise by a number of indicators.

The number of alleles for 15 microsatellite loci in the tested breeds ranged from 5 to 14, the average trait index being 7.53 ± 0.68 (Figure 1). In Holstein cattle, the greatest number of alleles was revealed at locus Tgla53 (14 alleles); the lowest number was at the Bm1824, Eth3, Ilst6, Csrn60, and Tgla126 (5 alleles) loci. The most variable loci were Tgla122, Tgla227, Csm66, and Tgla53; the number of alleles varied from 10 to 14.

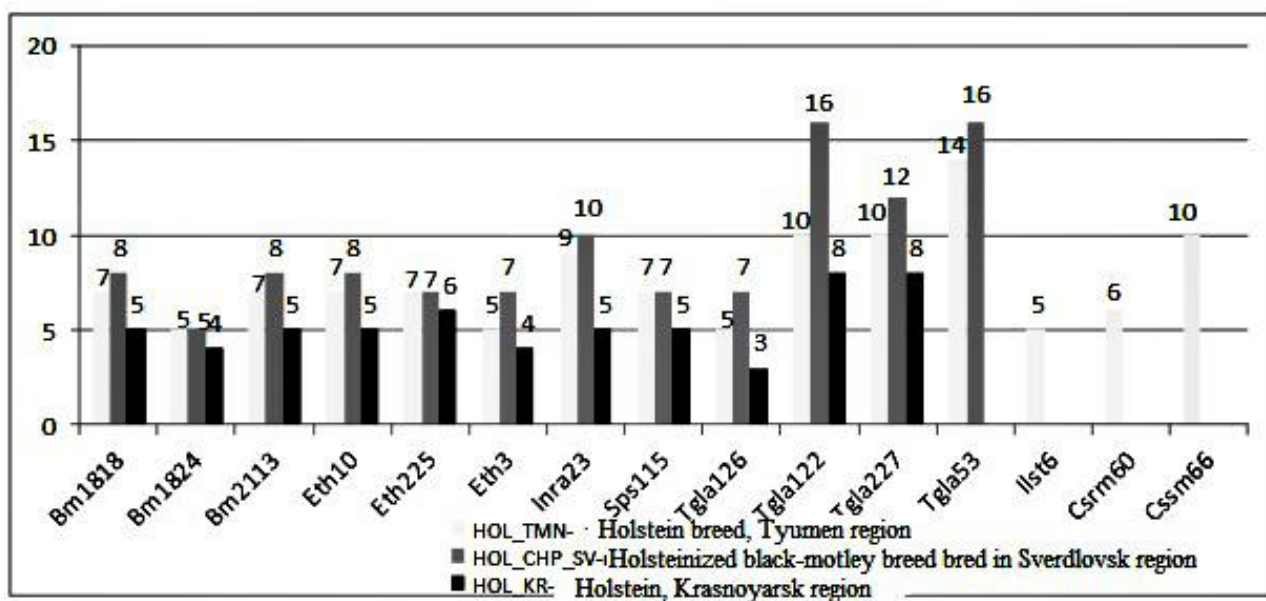


Figure 1. Number of alleles of microsatellite loci of dairy cattle under investigation at enterprises.

It can be assumed that the low values of the allele number for some loci (Bm1824, Tgla126) in the researched group of animals bred at the Tyumen Region enterprise are due to the use of semen obtained from bull producers belonging to a limited contingent, which can lead to a decrease in genetic diversity.

According to (3), Khabibrakhmanova, Kalashnikova (8), in populations of dairy cattle bred in Krasnoyarsk Krai and Sverdlovsk regions, the number of alleles for STR loci varied from 4 to 16. Comparison of the number of alleles at the loci under research in the populations of Tyumen and Sverdlovsk regions revealed differences by 1-2 alleles at the Bm181818,

Bm2113, Eth10, Eth3, Inra23, Tgla126, Tgla227, and Tgla53 loci.

No differences were found for Bm1824, Eth225, or Sps115 loci. It should be noted that in the populations of the Tyumen and Sverdlovsk regions, the most variable locus was Tgla126, with the number of alleles 14 and 16, respectively. In the Holstein breed of OAO Krasnoyarsk Agroplem, the number of alleles varied from 3 to 8, which is significantly lower compared to the above-mentioned populations (3, 8).

Table 2 shows the allele frequencies for 15

microsatellite loci in the animals under investigation, which turned out to be different. The maximum allele frequency of microsatellite loci allelic variants was revealed for 3 loci: Bm1818266 (0.52), Sps115248 (0.65), and Tgla126117 (0.63). The lowest allele frequency was detected at 17 loci: Eth10221, Eth225142, Inra23198, Inra23212, Inra23218, Sps115240, Tgla126119, Tgla122141, Tgla122173, Tgla53164, Tgla53176-178, Tgla22795, Tgla22799, Csm66177, Csm66197, Csm66199, and Ilst6296 with a range of 0.002 to 0.005.

Table 2. Frequency of 15 microsatellite loci alleles of Holstein breed animals bred at the enterprise in Tyumen region, Russia

Locus	Allele Allele frequency*	Number of alleles													
		1	2	3	4	5	6	7	8	9	10	11	12	13	14
Bm1818	allele	258	260	262	264	266	268	270							
	frequency	0,006	0,02	0,38	0,03	0,52	0,03	0,008							
Bm1824	allele	178	180	182	188	190									
	frequency	0,27	0,14	0,11	0,47	0,01									
Bm2113	allele	125	127	131	133	135	137	139							
	frequency	0,35	0,13	0,01	0,006	0,39	0,02	0,10							
Eth10	allele	209	213	217	219	221	223	225							
	frequency	0,10	0,06	0,21	0,45	0,004	0,07	0,10							
Eth225	allele	140	142	144	146	148	150	152							
	frequency	0,21	0,002	0,02	0,04	0,34	0,32	0,04							
Eth3	allele	117	119	125	127	129									
	frequency	0,41	0,10	0,09	0,15	0,34									
Inra23	allele	198	200	202	206	208	210	212	214	218					
	frequency	0,002	0,02	0,17	0,17	0,03	0,30	0,002	0,31	0,002					
Sps115	allele	240	248	252	254	256	258	260							
	frequency	0,002	0,65	0,15	0,04	0,11	0,008	0,05							
Tgla126	allele	115	117	119	121	123									
	frequency	0,31	0,63	0,002	0,03	0,03									
Tgla122	allele	141	143	149	151	159	161	163	171	173	183				
	frequency	0,002	0,41	0,19	0,10	0,008	0,07	0,12	0,04	0,002	0,06				
Tgla227	allele	81	83	87	89	91	93	95	97	99	103				
	frequency	0,01	0,02	0,07	0,36	0,09	0,01	0,002	0,39	0,002	0,05				
Tgla53	allele	154	158	160	162	164	166	168	170	172	174	176	178	186	188
	frequency	0,05	0,22	0,32	0,14	0,003	0,05	0,16	0,02	0,02	0,01	0,003	0,003	0,007	0,007
Csm66	allele	177	179	181	183	185	187	189	193	197	199				
	frequency	0,002	0,03	0,006	0,13	0,18	0,05	0,5	0,10	0,002	0,004				
Ilst6	allele	288	290	292	294	296									
	frequency	0,38	0,02	0,15	0,44	0,004									
Csm60	allele	92	96	98	100	102									
	frequency	0,23	0,18	0,10	0,08	0,41									

*Allele frequency in fractions of units.

For a number of the microsatellite loci under research in cattle, namely Bm1818²⁷⁰, Bm2113¹³³, Eth225¹⁴², Inra23¹⁹⁸, Inra23²¹⁸, Tgla126¹¹⁹, Tgla122¹⁴¹, Tgla122¹⁵⁹, Tgla122¹⁷³, Tgla127⁹⁵, Tgla127⁹⁹, Tgla53¹⁶⁴, Tgla53¹⁷⁶⁻¹⁸⁸, Tgla227⁹⁹, Csm66¹⁸¹, Csm66¹⁹⁷, and Csm66¹⁹⁹, unique allelic variants were identified that may be breed-specific markers for dairy animals.

When comparing the data on animals of the Tyumen region with the data available in the literature, the frequency of alleles for the investigated microsatellite markers turned out to be slightly lower compared to the population of the Holstein breed imported from Holland and Canada to OAO Krasnoyarsk Agroplem (Bm1818²⁶⁶ – 56%, Bm1824¹⁸⁸ – 60%, Eth10²¹⁹ – 60%, Sps115²⁴⁸ – 77%, Tgla126¹¹⁷ – 70%) (8).

On the basis of allelic frequencies for 15 microsatellite loci, the genetic diversity degree for animals at the intraspecific level was determined using the main quantitative indicators: the level of polymorphism (A_e), expected (H_e) and observed (H_o) heterozygosities, Wright's fixation index (F_{is}), and the informational polymorphism value (information content index, PIC). An objective algorithm for assessing polymorphism and formulas is well known (2, 7, 9, 10).

The genetic diversity analysis for the Holstein animals bred at the enterprise of the Tyumen region, Russia, according to the genetic variability indicators is presented in Table 3.

The level of polymorphism (A_e) was associated with the number of alleles in each investigated locus; with an increase in their number, the value of this indicator increased, making it possible to efficiently identify acting alleles in the presented sample. As a result of this study using STR markers, high values of the polymorphism level (A_e) were obtained at all loci within the range from 2.04 (Tgla126) to 5.02 (Tgla53). The average A_e value for the investigated sample was 3.31, which divided the animals into two equal groups. The first group included the following loci: Eth3, Csm66, Bm1818, Ilst6, Sps115, Bm1824, and Tgla126, which had an A_e value below the revealed average level. The second group of loci comprised Inra23, Tgla122, Eth225, Csm60, Eth10, Tgla53, and Tgla227; conversely, it was characterized by indicators higher than the revealed average for our sample, with the exception of the Bm2113 locus, which was equal to 3.32. In general, in the animal population under research, a high level of polymorphism was revealed for 7 investigated loci.

Table 3. The level of genetic variability of Holstein animals by microsatellite loci

Item No.	Locus	Holstein (n=241)				
		A_e	H_o	H_e	F_{is}	PIC
1	Eth3	3,18	0,91	0,69	-0,32	0,63
2	Csm66	3,20	0,77	0,69	-0,16	0,65
3	Inra23	4,06	0,78	0,76	-0,03	0,71
4	Bm1818	2,37	0,63	0,58	-0,09	0,50
5	Ilst6	2,72	0,63	0,63	0	0,56
6	Tgla122	4,17	0,80	0,76	-0,05	0,73
7	Sps115	2,19	0,59	0,54	-0,09	0,51
8	Eth225	3,50	0,79	0,72	-0,10	0,66
9	Csm60	3,72	0,87	0,73	-0,19	0,69
10	Bm2113	3,32	0,88	0,70	-0,26	0,65
11	Bm1824	3,08	0,74	0,68	-0,09	0,62
12	Eth10	3,67	0,78	0,73	-0,07	0,69
13	Tgla53	5,02	0,55	0,80	0,31	0,77
14	Tgla126*	2,04	0,46	0,51	0,10	0,43
15	Tgla227*	3,42	0,81	0,71	-0,14	0,66
	$\bar{X} \pm S_x$	3,31±0,20	0,73±0,03	0,68±0,02	x	0,63±0,02

*The sample of animals for loci Tgla126 and Tgla227 was n = 204.

To assess the genetic diversity in populations, it is necessary to take into account their heterozygosity, which is an important indicator for the genetic structure of organisms with a different composition of alleles of the same gene. The inheritance of different gametes by the descendants from the parents leads to the formation of heterozygotes. Heterozygosity is a tool of heterosis, in particular, in livestock breeding; it is used in the theory and practice of selecting/screening pairs to increase the productivity of the herd, and it also has a positive effect in the adaptation of organisms to changing environmental conditions; therefore, its assessment is an important criterion for calculating values in the population genetics (2).

The heterozygosity level observed reflected genetic variation in the population and was regulated by the frequency of heterozygotes. Expected heterozygosity (H_e) determines the expected level of precise genetic variation using allelic diversity for DNA markers in animals. The observed and expected heterozygosity values ranged from 0 to 1 (5). In general, a high heterozygosity level was revealed in the population under research. The highest level of observed heterozygosity (H_o), equal to 0.91, was found at the Eth3 locus. The lowest level of observed heterozygosity was found at the Tgl126 locus and amounted to 0.46. The expected heterozygosity level exceeded 0.51, which indicates high heterozygosity in all loci under research (7, 10).

The comparison made between the observed and expected heterozygosities for microsatellite loci revealed that the value of the average values of H_o and H_e heterozygosity in Holstein cattle bred in the Tyumen region do not differ significantly and are equal to 0.73 and 0.68, respectively. Thus, the close values of the H_o and H_e heterozygosity indices characterize a high level of genetic diversity in the sample of animals under research, reflect the competent selection and breeding work performed by the specialists of the Tyumen region enterprise, and indicate the low inbreeding of the herd.

According to the available literature data, this indicator in animals of the Sverdlovsk and Krasnoyarsk regions was approximately similar to those identified by us and amounted to 0.74 and 0.68, respectively (Table 4). It was revealed that the level of observed heterozygosity in animals from the samples of the Tyumen and Sverdlovsk regions was approximately the same with average values of 0.73 and 0.74, respectively; it turned out to be slightly lower in animals of the Krasnoyarsk Territory and amounted to 0.67 (3, 8).

The expected heterozygosity level in the Holsteinized black-and-white breed of the Sverdlovsk region was slightly higher than in the populations of the Tyumen and Krasnoyarsk regions, where it was approximately the same and amounted to 0.68 and 0.67, respectively.

Table 4. Indicators of genetic variability in dairy cattle

Region	Breed	Sample size, n	H_o	H_e	PIC	Literature sources
Tyumen region	Holstein	241	0,73	0,68	0,63	own research
Sverdlovsk region	Holstein black and white	404	0,74	0,73	0,69	(3)
Krasnoyarsk region	Holstein	24	0,67	0,67	0,61	(8)

To characterize the population level of variability, we used the Wright fixation index (F_{is}), which reflects the deviation of the frequencies of heterozygous genotypes from the theoretically expected proportion of heterozygotes and allows estimating the inbredness of the population during random mating within the population (5). The current study revealed that in 12 loci, the data obtained had negative values ranging from -0.03 to -0.32, indicating some excess of heterozygotes; only three loci, *Ilst6* (0), *Tgla126* (0.10), and *Tgla53* (0.31), showed their deficiency. Thus, the population under research is almost in Hardy-Weinberg equilibrium, with a slight deviation towards heterozygotes.

The informational polymorphism value (PIC) shows the ability of a marker to establish polymorphism in a population depending on the number of alleles detected and the distribution of their frequencies for each locus (5, 9). Values of this criterion close to 0 allow us to conclude that the population is monomorphic; vice versa, values close to 1 indicate its polymorphism. In addition, the locus is highly informative when the PIC is 0.5; when it is in the range of 0.25 to 0.5, it is rather informative; a value ≤ 0.25 means low informativity of the locus (11). As a result of the analysis of the data obtained, it was found that in the Holstein breed animals bred at the Tyumen region enterprise, all the microsatellite loci under research had a PIC value ≥ 0.50 . The highest PIC value was detected at the *Tgla53* locus (0.77) and the lowest at *Tgla126* (0.43). This

characterizes the high informativity level of the microsatellite markers used in genotyping. It should be noted that the information polymorphism value characterizes the diversity of allelic variants at the locus, which means that this index is suitable for estimating the informativity level in the group of animals under research.

The obtained results characterize the genetic variability of cattle bred in the Tyumen region and also agree with the results of studies of other scientists on the allelofund of the Holstein breed animals bred in the Sverdlovsk region. Thus, the loci used are applicable for assessing the genetic diversity of the animals bred in this enterprise of the Tyumen region, Russia.

It should also be noted that in breeding work, it is important for specialists of livestock enterprises to obtain reliable information on the origin of descendants by parents using microsatellite markers. Therefore, inaccuracies (errors) in the interpretation of fragment analysis data on STR loci of the cattle genome should be taken into account when testing animals.

As a result of our genotyping of animals in 120 incomplete (descendant/mother) families, 7% of erroneous entries in the pedigree of the descendants was found. Moreover, based on the interpretation of the data obtained on electrophoregrams at a number of loci (*Csm66*, *Eth3*), a number of artifacts were obtained in the form of additional peaks that made it difficult to recognize true peaks (Figure 2A) (10). True peaks that were located outside the binary matrix were identified (Figure 2B).

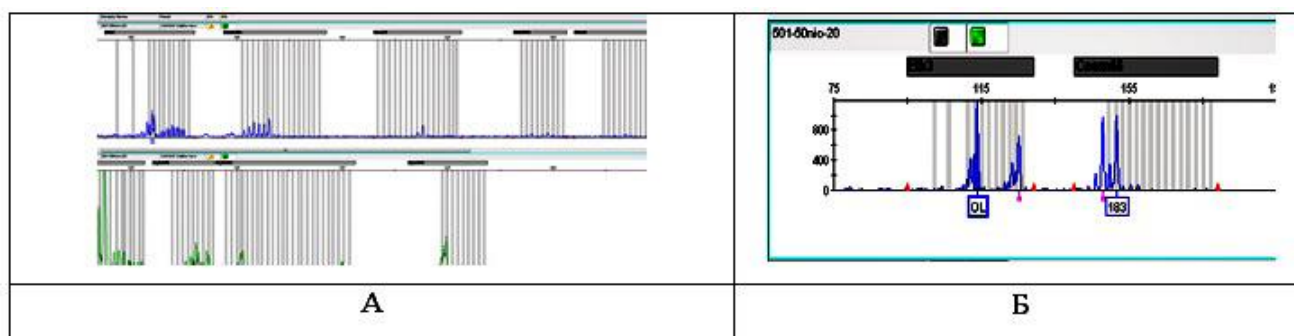


Figure 2. Data interpretation errors based on the results of microsatellite analysis of the genome of animals bred in the Tyumen region, Russia (additional (false) peak (A), peak outside the binary matrix (B)).

Thus, the use of the panel of 15 microsatellite loci made it possible to characterize the genetic structure of the cattle allelofund, assess genetic diversity, determine the breed affiliation of animals and the origin of descendants by parents with a high degree of reliability of the results, as well as to use individual loci more effectively for various studies in genetics. The high level of polymorphism at the loci under research made it possible to study the genetic variability level for the cattle and opened the possibility of searching for new relationships between allelic variants of microsatellite loci with productive traits of animals.

4. Conclusion

The following conclusions can be made based on the results obtained:

1) The allelic diversity and characterization of the genetic structure for Holstein breed cattle bred at the enterprise in Tyumen region in 241 head by 15 microsatellite loci such as Eth3, Csm66, Inra23, Bm1818, Ilst6, Tgla122, Sps115, Eth225, Csm60, Bm2113, and Bm1824 were analyzed. Eighteen unique allelic variants at 10 microsatellite loci that can be used as breed-specific markers for genetic identification of dairy cattle were revealed.

2) A high genetic variability level of dairy cattle bred in the Tyumen region was revealed. A high level of polymorphism (A_e 3.32) was detected in 47% of the loci under research. The maximum values of the observed heterozygosity (H_o) for the Eth3 locus (0,91) and the value of informational polymorphism (PIC) for the Tgla53 locus (0,77) were revealed.

Herein, 7% of erroneous entries in the pedigree of descendants in 241 animals of Holstein breed bred in the Tyumen region were revealed. The results of experiments on DNA testing of animals using microsatellite loci of the cattle genome were obtained. The Csm66 and Eth3 loci revealed true and additional peaks that distorted the true value of the results.

Authors' Contribution

Kabitskaya Ya.A. designed and performed all experiments, and wrote the first draft of a manuscript, designed the statistical models with based science articles (Glinskaya N.A., Peculiarities of SSR polymorphism in horses / N.A. Glin-skaya, E.I. Prilovskaya, D.A. Kaspirovich, O.A. Epishko, E.S. Cheburanova // Bulletin of Polesky State University. 2017. -No. 1. - P. 8-13.; Chesnokov, Yu.V., Estimation of the measure of information polymorphism of genetic diversity / Yu.V. Chesnokov, A.M. Artemyeva // Agricultural Biol-ogy. 2015. - Vol. 50. -No. 5. - P. 571-578. Merkuryeva E.K. Genetic basis of breeding in animal husbandry: a training manual / E.K. Merkuryeva. M.: Kolos. 1977. - 174 p., Nei M., Sampling variances of heterozygosity and genetic distance / M. Nei, A.K. Roychoudhury // Genetics. 1974. 76:379 – 390.) Kabitskaya Ya.A. ana

lyzed the data. Boyko E.G. verified the analytical methods, and Boyko E.G. proofread and revised the final version of the present manuscript. Kabitskaya Ya.A. performed radioimmunoassays.

Ethics

The present study was approved by the Ethics Committee of the Ministry of Agriculture of the Russian Federation.

Conflict of Interest

The authors declare that they have no conflict of interest.

Acknowledgment

The current work was carried out within the State Assignment of the Ministry of Agriculture of the Russian Federation: No.082-03-2020-259 (AAAA4-A20-120042990035-9) and R&D project AAAA-A20-120120490045-8.

References

1. Gladyr E, Gorelov P, Chinarov I, Zinov'eva N. Evaluation of test system performance based on

- microsatellites in DNA-examination of cattle. *Achiev Sci Technol Agroindustrial Complex*. 2011;8(1):51-4.
2. Glinskaya N, Prilovskaya E, Kaspirovich D, Epishko O, Cheburanova E. Peculiarities of SSR polymorphism in horses. 2017;5(1):8-13.
 3. Modorov M, Tkachenko I, Green A. Genetic variability of Holsteinized black-motley cattle in the Sverdlovsk region. *Problems of normative-legal regulation in veterinary medicine*. 2019;4(1):119-22.
 4. Sambrook J, Fritsch E, Maniatis T. *Molecular cloning a laboratory*. Cold Spring Harb. 1989;7:37-9.
 5. Chesnokov Y, Artemyeva A. Estimation of the measure of information polymorphism of genetic diversity. *Agric Biol*. 2015;50(5):571-8.
 6. Merkuryeva E, Kolos M. Genetic basis of breeding in animal husbandry: a training manual. *Eur Sci Rev*. 1997;5(2):174.
 7. Nei M, Roychoudhury AK. Sampling variances of heterozygosity and genetic distance. *Genetics*. 1974;76(2):379-90.
 8. Khabibrakhmanova Y, Kalashnikova L, Golubkov A, Lefler T, Golubkov A, Mirvaliev F. Genetic polymorphism of Holstein bulls of JSC "Krasnoyarskagroplem" on the basis of microsatellite DNA markers. *Bulletin of KrasGAU*. 2019;3(1):135-40.
 9. Botstein D, White R, Scolnick M, Davis R. Construction of a Genetic linkage map in man using restriction fragment length polymorphisms. *Am J Hum Genet*. 1980;32(1):314-31.
 10. Kabitskaya Y, Kalashnikova L, Boyko E, Kalashnikov A. Genetic identification as a criterion for coincidence with the data of primary accounting of animals on the territory of UFO. *Bulletin of RSATU*. 2020;1(1):114-21.
 11. Kuznetsov V. Wright's F-statistics: evaluation and interpretation. *Problem Biol productive Anim*. 2014;4(1):80-104.