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Use of the Molecular-Genetic Markers in the Selection Process of the Ukrainian Animal Husbandry

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Aim. To analyze the genetic structure of the Holstein and Simmental breeding bulls from various breeding stock-rearing farms of Ukraine by polymorphisms of kappa-casein, β-lactoglobulin, growth hormone, leptin, pituitary-specific transcription factor, myostatin loci genes. **Methods.** Individual genotypes of the animals were determined using the polymerase chain reaction (PCR) followed by determination of restriction fragment length polymorphism (RFLP analysis). **Results**. A complex model genotype for increasing milk yield of the animals of the Ukrainian Black and White Dairy breed – κ-Cn^{AB}, βLG^{AB}, GH^{LV}, Pit-1^{AA} LEP^{AA}, butterfat content – κ-Cn^{AB}, βLG^{AB}, GH^{VV}, Pit-1^{AA}, LEP$^{BB/AB}$; Ukrainian Red and White Dairy breed – $\dot{C}$n^{AA}, βLG^{AA}, GH^{L}, PIt-1^{AB} and κ-Cn^{AA}, βLG^{AB}, GH^{LV}, Pit-1^{AB}; Simmental breed – κ-Cn^{BB}, βLG^{BB}, GH^{LL}, LEP^{AB} and κ-Cn^{BB}, βLG^{BB}, GH^{LV}, LEP^{AA} was determined respectively. Genetic certification of the breeding bulls of 25 various breeds on standard microsatellite panels (BM1824, BM2113, INRA023, SPS115, TGLA122, TGLA126, TGLA227, ETH10, ETH225, ETH3) was held for their biological material long-term storage rationale in the National Bank of Genetic Resources. **Conclusions.** Comprehensive monitoring of cattle breeding resources predetermines the implementation of genetic examination of the breeding animals' origin according to ISAG international guidelines in Ukraine. This will determine the specific type or breed gene pool as a whole, also characterize the vector of micro-evolutionary processes in the populations of animals and allow to solve other breeding and genetic problems.

Key words: cattle, DNA polymorphism, quantitative traits loci (QTL), genotype, microsatellite (STR).

INTRODUCTION

It is customary to consider the genetic potential of the livestock in terms of the possibility to form the gene sets able to determine a phenotype in certain environmental conditions.

Having the detailed information on closely linked genes, the knowledge of genetics of quantitative traits nowadays provides the molecular genetic marking of "major" genes of quantitative traits, and thus, allows to predict and get genotypes of newborn animals endued with desirable phenotypic traits.

Genetic markers make it possible to obtain information about features of breeding material, assess the diversity of gene pool of farm animals, predict changes associated with breeding factors, identify potentially high-productive animals at an early age, aiming to make selection of breeding pairs per genotype for heterotic effect in their offspring [1, 2]. Molecular genetic monitoring of populations allows to control their genetic structure for maintaining optimal balance complex of alleles and to analyze the genotype of the animals at the level of genes associated with economically useful traits. These genes belong to the so-called quantitative traits loci, QTL [3−5].

Identification of genes and their mutations, which determine direction and extent of QTL development both in the Europe and US, provides large profits by reducing the generation interval, early introduction of breeding stock in reproduction and using selection with markers (MAS) [6].

In order to intensify the selection process in the cattle breeding of Ukraine, the implementation of new methods and approaches based on the analysis of DNA polymorphism exactly is an important and priority task of the modern agricultural science.

MATERIALS AND METHODS

The blood samples of the Ukrainian Black and White Dairy breed (125 animals), Ukrainian Red and White Dairy breed (90 animals), Simmental breed (112 animals) and Holstein breed (53 animals) cows – 380 animals in total – from various farms, including the breeding stock-rearing ones, all over Ukraine: Chrystynivske Enterprise Research Farm, Progress Joint Stock Company and Scientific Production Association, Genetic Resources Limited Liability Corporation, Agro-Region Private Joint Stock Company, Agro Firm Kyivska Limited Liability Corporation from the Kiev region, Menske Breeding Livestock Enterprise Joint Stock Company from the Chernihiv region, Khmelnytske General Breeding Livestock Enterprise Joint Stock Company, Viytovetske Breeding Livestock Enterprise Joint Stock Company from the Khmelnytsky region; DNA Bank of Animal Genetic Resources of the Institute of Animal Breeding and Genetics of the National Academy of Agricultural Sciences were used in course of the present research.

The genome DNA was isolated from peripheral blood of the animals using the standard commercial set "DNA-Sorb B" (Amplysens, Russian Federation) according to the manufacturer's recommendations.

Table 1. Methodological Features of PCR-RFLP Analysis QTL In the Cattle

Locus	Amplified fragment length, bp	Restriction enzymes	Restriction products length, bp
κ-Cn	273	Hinf 1/Hind III	273, 224, 133, 91, 40 273, 182, 91
βLG	247	Hae III	247, 148, 99, 74
GH	223	AluI	223, 171, 52
LEP	1830	Sau3AI	1830, 740, 690, 470, 400, 310, 220, 90
PIT-1	1355	HinfI	1355, 660, 425, 385, 270, 40
MSTN	196	–	–

Table 2. Sequence of Primers For STR-Analysis

Chromosome	Length, bp	Motif	Primer (5'-3')
BM1824 (D1S34)	170–218	$(GT)_n$	F* – GAG CAA GGT GTT TTT CCA ATC R** – CAT TCT CCA ACT GCT TCC TTG
BM2113 (D2S26)	114–146	$(CA)_n$	F – GCT GCC TTC TAC CAA ATA CCC R – CTT CCT GAG AGA AGC AAC ACC
INRA023 (D3S10)	193–235	$(AC)_n$	F – GAG TAG AGC TAC AAG ATA AAC TTC R – TAA CTA CAG GGT GTT AGA TGA ACT C
SPS115 (D15S21)	235–265	$(CA)_n TA (CA)_6$	F – AAA GTG ACA CAA CAG CTT CTC CAG R – AAC GAG TGT CCT AGT TTG GCT GTG
TGLA122 (D21S6)	134–193	$(AC)_n(AT)_n$	F – AAT CAC ATG GCA AAT AAG TAC ATA C R – CCC TCC TCC AGG TAA ATC AGC
TGLA126 (D20S1)	104–131	$(TG)_n$	F – CTA ATT TAG AAT GAG AGA GGC TTC T R – TTG GTC TCT ATT CTC TGA ATA TTC C
TGLA227 (D18S1)	64–115	$(TG)_n$	F – CGA ATT CCA AAT CTG TTA ATT TGC T R – ACA GAC AGA AAC TCA ATG AAA GCA
ETH10 (D5S3)	198–234	$(AC)_n$	F – GTT CAG GAC TGG CCC TGC TAA CA R – CCT CCA GCC CAC TTT CTC TTC TC
ETH225 (D9S1)	135–165	$(TG)_4 CG (TG)(CA)_n$	F – GAT CAC CTT GCC ACT ATT TCC T R – ACA TGA CAG CCA GCT GCT ACT
ETH 3 D19S2	90–135	$(GT)_n AC (GT)_6$	F – GAACCTGCCTCTCCTGCATTGG R – ACTCTGCCTGTGGCCAAGTAGG

Note. *F – forward; **R – reverse.

The restricted fragments' length polymorphism of the genes under exploration was PRC-RFLP analyzed. The following reaction mixture, total volume of 10 µl was used for PCR: diH_2O – 4.3 µl, 5-x PCR-buffer (15 M Mg – 1.0 mol) – 2.0 µl; 10 dNTP mixture (concentration – 2 mM each) – 0.8 µl; 2 primers (70 ng each) – 0.8 µl; Taq-polymerase (1 mol/1000 U) – 0.1 µl; DNA (50−100 ng) – 2.0 µl.

Both the temperature regime and cycle number of PCR-amplification were determined for each of the genes separately. The studied genes' fragments were amplified applying the following primers: for the kappa-casein locus, κ-Cn: (5′-GAAATCCCTACCATCAATACC-3′ and 5′-CCATCTACCTAGTTTAGATG-3′); β-lactoglobulin, βLG: (5′-GTGCTGGACACCGACTACAAAAAG-3′ and 5′-GCTCCCGGTATATG-ACCACCCTCT-3′) [7]; growth hormone, GH: (5′-GCTGCTCCTGAGGGCCCTTC-3′ and 5′-GCGGCGGCACTTCATGACCC-3′) [8]; leptin, LEP: (5′-GTCACCAGGATCAATGACAT-3′; and 5′-AGCCCAGGAATGAAGTCCAA-3′) [9]; pituitary-specific transcription factor, PIT-1: (5′-CAATGAGAAAGTTGGTGC-3′ and 5′-TCTGCATTCGAGATGCTC-3′) [10]; myostatin, MSTN: (5′-TCTAGGAGAGATTTTGGGCTT-3′ and 5′-TGGGTATGAGGATACTTTTGC-3′) [11]. The following restriction enzymes selected for each locus were used to analyze the κ-Cn, βLG, GH, PIT-1, LEP genes' polymorphism (Table 1).

Table 3. The Genetic Structure of Studied Breeds by Quantitative Traits Loci

Locus	Ukrainian Black and White Dairy (n = 125)		Ukrainian Red and White Dairy (n = 90)		Holstein (n = 53)		Simmental (n = 112)	
	Frequency							
	Genotype	Allele	Genotype	Allele	Genotype	Allele	Genotype	Allele
κ-Cn	$\kappa\text{-Cn}^{AA}$ 0.664	A 0.820	0.778	A 0.888	0.793	A 0.896	0.467	A 0.679
	$\kappa\text{-Cn}^{AB}$ 0.312	B 0.180	0.222	B 0.112	0.207	B 0.104	0.425	B 0.320
	$\kappa\text{-Cn}^{BB}$ 0.024		0.000		0.000		0.108	
βLG	βLG^{AA} 0.08	A 0.368	0.078	A 0.367	0.132	A 0.415	0.152	A 0.397
	βLG^{AB} 0.576	B 0.632	0.578	B 0.633	0.566	B 0.585	0.489	B 0.603
	βLG^{BB} 0.344		0.344		0.302		0.359	
GH	GH^{LL} 0.616	L 0.780	0.833	L 0.917	0.793	L 0.896	0.402	L0.598
	GH^{LV} 0.328	V 0.220	0.167	V 0.083	0.207	V 0.104	0.391	V0.402
	GH^{VV} 0.056		0.000		0.000		0.207	
PIT 1	$PIT1^{AA}$ 0.192	A 0.436	0.155	A 0.438	0.208	A 0.416	0.000	A 0.141
	$PIT\ 1^{AB}$ 0.488	B 0.564	0.567	B 0.562	0.417	B 0.584	0.281	B 0.859
	$PIT\ 1^{BB}$ 0.320		0.278		0.375		0.719	
LEP	LEP^{AA} 0.616	A 0.788	–	–	–		0.598	A 0.799
	LEP^{AB} 0.344	B 0.212	–	–	–		0.402	B 0.201
	LEP^{BB} 0.04		–	–	–		0.000	

Restriction products were separated by electrophoresis in 2 per cent agarose gel followed by their ethidium bromide staining. Visualization was performed on UV light transilluminator followed by digital photographing of the electrophoregramms. The amplicon size differentiation was performed using molecular weight marker GeneRuler TM 50 bp DNA Ladder, # SM0378, (Fermentas, Lithuania).

The STR-markers analysis of various cattle breeds' genetic structure (Table 2) was performed applying the ABI Prism 3130 Genetic Analyzer (Applied Biosystems, USA).

Genetic population and biometric analyses of the obtained results were performed using the mathematical statistics procedures applying the standard software GenAlex6, BIOSIS-1, Statistica.

RESULTS AND DISCUSSION

The obtained results that concern determination of the genetic structure of four above mentioned cattle breeds for κ-Cn, βLG, GH, LEP, PIT-1 and MSTN loci allowed to perform the comparative analysis of their gene pool as for the explored genes (Table 3).

AA genotype frequency for κ-Cn locus is 0.664, 0.778 and 0.793 for the Ukrainian Black and White Dairy, Ukrainian Red and White Dairy and Holstein breeds respectively, while the A-allele homozygous animals demonstrate frequency of 0.467 for the Simmental breed.

There were no BB-genotype homozygous animals detected in the Holstein and Ukrainian Red and White Dairy breeds, while the frequency of the animals of the kind for the Ukrainian Black and White Dairy breed is 0.024, that is 4.5 times less than in the population of the dual-purpose Simmental breed demonstrating 0.108. From the point of view of the allele assortment, the Ukrainian Black and White Dairy, Ukrainian Red and White Dairy and Holstein breeds were proven to be the likest, while the A-allele frequency is 0.820, 0.888 and 0.896 respectively. The Simmental breed animals demonstrated significantly lower frequency for this very allele – 0.679.

For the milk breeds, the correlation between the B-allele frequency decrease and milking capacity raise was stated as for the κ-Cn locus. The B-allele frequency for the Ukrainian Black and White Dairy breed is 0.180, Ukrainian Red and White Dairy – 0.112, Holstein – 0.104, while only 0.320 for the Simmental breed ($p < 0.01$).

As for βLG locus, the AB-genotype animals prevail among those surveyed. The probability of difference between actual genotype assortment and the predicted one for the Ukrainian Black and White Dairy and Ukrainian Red and White Dairy breeds is statistically essential ($p < 0.01$). This genotype frequency for the two above mentioned breeds is almost the same: 0.576 and 0.578 respectively. It is also very like to the Holstein animals result – 0.566, whereas somewhat less for the Simmental breed – 0.489. The BB-genotype homozygous animals frequency is equal for the Ukrainian Black and White Dairy and Ukrainian Red and White Dairy breeds – 0.344, while 0.302 for the Holstein breed and 0.359 for the Simmental animals.

The AA-genotype frequency, as for βLG locus, is 0.152 for the Simmental breed, while for the Ukrainian Black and White Dairy and Ukrainian Red and White Dairy breeds – 0.08 and 0.078 respectively, it means much less. At the same time this frequency is 0.132 for the Holstein animals.

No GH^{VV} homozygous animals were detected in the populations of the Ukrainian Red and White Dairy and Holstein breeds, while the portion was negligible in the Ukrainian Black and White Dairy population – 0.056 only. Finally, this desirable genotype frequency for the Simmental breed is 0.207. The GH^{LV} heterozygous genotype was following: the Ukrainian Black and White Dairy breed – 0.328, Ukrainian Red and White Dairy – 0.167, Holstein – 0.207 and Simmental – 0.391. The high frequency of GH^{LL} homozygotes appeared to be specific for the milk breeds as compared to the Simmental animals demonstrating this genotype frequency at 0.402, while for the Ukrainian Black and White Dairy, Ukrainian Red and White Dairy and Holstein breeds this value was 0.616, 0.833 and 0.793 respectively. L-allele frequency was the least for the Simmental animals – 0.598. The likeness of the genetic structure was also stated for the studied livestock milk breeds as for this very locus as well.

As for PIT-1 locus, the BB homozygotes' higher frequency is peculiar for the Simmental animals as compared to other breeds – 0.719, while no A-allele homozygous animals were detected, and the AB heterozygous ones' frequency was 0.281. Both the genotype and allele variants frequency distribution for the Ukrainian Black and White Dairy, Ukrainian Red and White Dairy and Holstein breeds were analogous.

The AA-genotype frequency for the Ukrainian Black and White Dairy, Ukrainian Red and White Dairy and Holstein animals was 0.192, 0.155 and 0.208 respectively. The B-allele homozygotes were distributed as follows: the Ukrainian Black and White Dairy breed –

0.320, the Ukrainian Red and White Dairy breed – 0.278 and the Holstein one – 0.375. Contrast to the others, the Simmental breed is distinguished by high frequency of the BB-homozygotes as for the PIT-1 locus – 0.719, whereas A-allele homozygous animals were never detected. The A-allele frequency for the Ukrainian Black and White Dairy, Ukrainian Red and White Dairy and Holstein breeds was 0.436, 0.438 and 0.416 respectively. This has been caused by both the breeding aim and the native breeds history.

The genotype distribution as for the LEP locus was for the Ukrainian Black and White Dairy breed the following: AA – 0.616, AB – 0.344, BB – 0.04; for the Simmental breed: AA – 0.598, AB – 0.402, while no BB-homozygous species were detected. The A-allele frequency for the Ukrainian Black and White Dairy breed was 0.788, for the Simmental one – 0.799.

In general, the explored QTL demonstrated the insufficient heterogeneity rate within the compared populations as concerning the population genetic characteristics. Like that, the homozygosity index (Ca) was within 0.508 in the Ukrainian Black and White Dairy breed (for the PIT-1 locus) and 0.848 in the Ukrainian Red and White Dairy one (for the GH locus). The allele effective quantity (Ne) that reflects the polymorphism rate for each diallel locus varied inefficiently: from 1.179 in the Ukrainian Red and White Dairy breed to 1.969 in the Ukrainian Black and White Dairy cows. Quite a notable variability rate was fixed for the Ukrainian Red and White Dairy animals, while the effective heterozygosis rate (H_o) varied from 0.167 (for the growth hormone gene) to 0.578 (for the βLG locus). This could give evidence on existence of some genetically automatic processes connected to the “blood refreshing” in the Holstein breed. In whole, the expected heterozygosis appeared to be higher than the actual one for all explored breeds.

The accumulation of the heterozygous genotypes in the livestock populations could be caused by the usage of the breeders with the corresponding genes’ sets. The involvement of the heterozygous species into the selective process may be also possible, whereas these ones are usually better adapted for the maintenance technologies and also provides higher performance. The fixation index (F) values for various breeds should be noted apart. So, -0.003 in the Ukrainian Black and White Dairy could give evidence on the selective neutrality of this very locus, *i. e*. the genotypes’ dispersal as for this very locus never diverges from the potentially expected after the Hardy-Weinberg principle ($\chi^2 = 0.405$). On the contrary, the fixation index value in the Simmental breed as for the growth hormone gene is +0.395, what can be caused by the homogeneous intra population selection resulted in increase of the homozygous genotypes’ amount. As concerning the GH locus, it causes the raise of the VV genotypes’ concentration, which are associated with the higher butterfat percentage peculiar to the Simmental, as a dual-purpose breed only.

Differentiation between animals of various breeds as for correlations between milk yielding and certain genotype for κ-Cn locus allowed to identify various associations such as "genotype – character" depending on the breed. Thus, the milk yield of the Ukrainian Black and White Dairy AB-cows exceeds the AA-cows by 18.9 per cent ($p < 0.1$), AB > BB – by 24.9 per cent ($p < 0.05$); as for the Ukrainian Red and White Dairy breed: AA > BB – by 8 per cent; as for the Simmental livestock: BB > AA – by 4.09 per cent, BB > AB – by 14.7 per cent ($p < 0.1$).

The comparative analysis of milk performance of various cattle breeds depending on the genotype, as for βLG locus, provided with the following results: the protein content in milk of the Ukrainian Black and White Dairy AB-cows exceeds the result of the BB-species by 0.13 per cent, while AB > AA by 1.18 per cent ($p < 0.01$); as for the milk yield of the Ukrainian Red and White Dairy breed: AA > AB – by 8.5 per cent ($p < 0.05$), AA > BB – by 9.5 per cent ($p < 0,1$), the same breed, as for the fat content in milk: AB > – AA by 0.16 per cent and AB > BB – by 0.08 per cent; as for the fat content in milk in the Simmental animals: BB > AB – by 0.04 per cent.

For the GH growth hormone locus, the milk yield in the Ukrainian Black and White Dairy LV-cows exceeds the LL-ones’ result by 10.9 per cent ($p < 0.1$), LV > VV – by 7.3 per cent, as for the fat content in milk: LL > LV – by 0.038 per cent, VV > LL – by 0.037 per cent, VV > LV – by 0.07 per cent; in the Ukrainian Red and White Dairy cows, for the protein content in milk: LL > LV – by 0.001 per cent, for fat content in milk: LV > LL – by 0.099 per cent; in the Simmental animals, as for the milk yield: LL > LV – by 2.7 per cent and LL > VV – by 10.4 per cent, as for the fat content in milk: LV > LL – by 0.23 per cent and LV > VV – by 0.24 per cent. The increase in concentration of L-allele among populations indicates its selective advantage over the V-variant because of its greater milk performance effect.

As for PIT-1 locus, such a characteristic as milk yield appeared to be higher in the Ukrainian Black and White Dairy AA-cows for 8.9 per cent than in the BB-species,

AA > AB – by 6.1 per cent; the milk yield in the Ukrainian Red and White Dairy breed: AB > AA – by 4.8 per cent and AB > BB – by 4.3 per cent; the protein content in milk: AB > AA – by 0.021 per cent; the fat content in milk: AB > AA – by 0.064 per cent and AB > BB – by 0.002 per cent.

For leptin LEP gene, the Ukrainian Black and White Dairy animals showed highly probable difference as for fat content in milk: BB > AB – by 0.06 per cent ($p < 0.01$) and BB > AA – by 0.5 per cent ($p < 0.01$), AB > AA – by 0.4 per cent ($p < 0.01$); the Simmental breed as for milk yield: AB > AA – by 0.31 per cent, and as for the protein content in milk: AB > BB – by 0.05 per cent. The diverse nature of correlations between the leptin LEP gene and animals' pedigree should be noted. Thus, the high fat content in milk in the Ukrainian Black and White Dairy cows was caused by the presence of BB- and AB-species.

The animal genotype determination method by 10 microsatellite loci have been tested and modified (Table 4).

Generally speaking, for microsatellite locus TGLA126, nine allele variants were found among the surveyed animals, their size ranged from 114 to 130 bp. The largest number of alleles (nine) were observed in the Holstein breed, and the smallest (two) – in the Gascony, Shvitska, Pinzgau and Limousine. Allele sizes of 114 and 130 bp were typical only for the Holstein breed.

For microsatellite locus TGLA122, 22 allele variants ranging in size from 142 to 190 bp were received. The largest number of alleles of various lengths were observed in the Holstein breed (eighteen), and the smallest (one) – in the animals of the Shvitska and Maine-Anjou breeds. The animals of the Brown Carpathian breed have alleles of size 164 and 172 bp, which have

Table 4. The Total Number of Microsatellite Loci Allele Variants In Various Livestock Breeds

Breed	Microsatellite locus									
	1	*2*	*3*	*4*	*5*	*6*	*7*	*8*	*9*	*10*
Holstein	9	18	12	9	10	13	12	9	9	8
URD	4	7	7	6	5	5	7	5	5	3
URaWD	7	10	11	6	8	10	8	10	8	8
UBaWD	3	6	4	4	5	4	4	4	4	2
Angler	4	7	7	7	8	5	6	5	3	5
Simmental	7	9	9	6	7	7	10	8	4	8
Brown Carpathian	7	8	8	7	9	6	9	8	7	6
Lebedynska	4	5	5	6	6	4	7	6	4	5
Bald Ukrainian	4	7	8	5	6	5	7	5	4	6
Grey Ukrainian	4	6	3	5	6	5	7	4	4	7
Red Steppe	3	3	4	4	3	3	3	4	3	2
Jersey	3	2	3	1	2	2	3	2	3	3
Gascony	2	2	1	1	1	2	2	2	2	2
Shvitska	2	1	2	1	2	2	2	2	2	1
Pinzgau	2	4	4	3	4	5	5	4	3	2
Volyn Beef	5	10	10	5	6	5	7	8	6	7
Ukrainian Beef	6	7	10	5	6	7	12	8	4	8
South Beef	4	4	3	4	2	3	4	4	2	3
ZtPB	4	5	4	3	4	3	5	4	1	2
Charolais	5	4	5	4	5	4	6	6	1	3
Limousine	2	3	5	3	2	2	3	3	3	3
Kian	2	4	4	4	3	2	6	6	2	4
Maine-Anjou	3	1	2	2	2	1	2	2	2	2
Light Aquitaine	3	6	4	3	3	3	4	3	3	5

Comment: URD – Ukrainian Red Dairy breed; URaWD – Ukrainian Red and White Dairy breed; UBaWD – Ukrainian Black and White Dairy breed; ZtPB – Znamiansky type of Polissia Beef breed
1 – TGLA126; *2* – TGLA122; *3* – INRA023; *4* – ETH3, *5* – ETH225, *6* – BM1824, *7* – TGLA227; *8* – BM2113, *9* – ETH10, *10* – SPS115

never been met in any other species. Only animals of the Gray Ukrainian breed possessed the allele of size of 186 bp, while the animals of the Volyn and Southern Beef breeds had the allele variant in size of 184 bp.

For INRA023 locus, 14 allele variants ranging from 208 to 234 bp were revealed. The allele variant in size of 234 bp was fixed only in the animals of the Pinzgau breed.

Among the breeds studied for ETH3 locus allele size ranged from 108 to 134 bp. The animals of the Holstein breed possessed the allele variant in size of 134 bp, and the Ukrainian Red Dairy animals – 108 bp, which is peculiar only for this very breed. The allele variant of 116 bp was fixed in the bulls of the Limousin and Gray Ukrainian breeds, while the variant of 112 bp – in the animals of the Kian and Limousine breeds. In general, 13 allele different-size variants were revealed for this locus.

For ETH225 locus, 13 allele variants in size 140 to 164 bp were detected. For the breeders of the Angler and Ukrainian Red Dairy breeds the allele in size of 158 bp is typical, while allele variants in size of 160 bp – for the animals of the Lebedynska breed, 162 bp – for the Holstein breed and 164 bp – for the Brown Carpathian breed.

The allele variant in size of 198 bp, for BM1824 locus, was observed only in the animals of the Angler and Pinzgau breeds. In total, 14 allele variants within the range 176 to 202 bp were fixed.

The microsatellite locus of TGLA227 was represented by 14 allele variants different in size, ranging from 80 to 104 bp. The allele variant in size of 72 bp was revealed in the Ukrainian Beef breeders, whereas the one of 78 bp – in the animals of the Simmental and Ukrainian Beef breeds.

BM2113 locus has 11 different-size allele variants – 114 to 142 bp. The allele of 114 bp is typical for the Holstein breed, while the allele variants in size of 140 bp – for the Volyn Beef, Ukrainian Beef and Kian breeders.

Ten allele variants ranging in size from 212 to 232 bp were revealed for the microsatellite locus of ETH10. The allele of 232 bp was observed in the animals of the Ukrainian Red and White Dairy breed, 212 bp – in breeders of the Pinzgau breed.

For microsatellite locus SPS115, 11 alleles within the wavelength range 244 to 264 bp were revealed. The variant of 264 bp is typical for the animals of the Ukrainian Red and White Dairy breed.

Keeping in mind the results obtained from the explorations performed, it was conceded that the QTL alleles' influence upon the expression of the genes responsible for some lactation peculiarities changes within various breeds, while each population creates not only the unique gene pool, but also the original gene set. It depends on the substantial development of the multigenic characters used in the livestock selection and stock breeding owing to the maintenance peculiarities. The QTL genetic monitoring of the livestock populations together with the breeding value estimation methods allow to perform targeted gene selection and also identify individuals possessing prominent genetic potential among the cattle herds.

CONCLUSIONS

The results of the genetic population explorations involved three milk and one dual-purpose breeds of cattle give evidence on the promising outlook of the populations' heterogeneity rate estimation by examined QTL loci within peculiar herds in course of selection pressure determination.

Livestock genotypes and alleles' distribution together with their development are driven by the peculiarities of selection accomplished as for each se-parate breed depending on the performance purpose, though never connected to inbreeding during reproduction of the animals and their belonging to one or several lines. Alongside to the standard selection and stock breeding methods, the obtained data allow to shape the animals' populations owing to targeted genetic selection and parental combination possessing the necessary genetic potential as for milk yield, in particular.

The microsatellite loci analysis of the genetic structure peculiarities resulted in breeding bulls' genotypes for various livestock breeds. The obtained data give grounds to advise the corresponding panel of STR-markers for genotype determination and livestock origin validity control according to ISAG international guidelines.

Використання молекулярно-генетичних маркерів у селекційному процесі тваринництва України

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Мета. Аналіз генетичної структури бугаїв голштинської і симентальської порід різних племпідприємств України за поліморфізмом генів капа-казеїну, β-лактоглобуліну, гормону росту, лептину, гіпофізарно-специфічного фактора транскрипції, міостатину. **Методи.** Індивідуальні генотипи тварин ідентифікували за допомогою полімеразно-ланцюгової реакції (ПЛР) з подальшим визна-

ченням поліморфізму довжин рестрикційних фрагментів (ПДРФ-аналіз). **Результати.** Встановлено комплексний модельний генотип для підвищення надою тварин української чорно-рябої молочної породи – к-Cn^{AB}βLG^{AB} GH^{LV}Pit-1^{AA}LEP^{AA}, жирномолочності – к-Cn^{AB}βLG^{AB} GH^{VV}Pit-1^{AA}LEP$^{BB/AB}$; української червоно-рябої молочної породи – к-Cn^{AA}βLG$^{AA}GH^{LL}$PIt-1^{AB} та к-Cn^{AA}βLG^{AB} GH^{LV}Pit-1^{AB}; симентальської породи – к-Cn^{BB}βLG^{BB} GH^{LL}LEP^{AB} та к-Cn^{BB}βLG$^{BB}GH^{LV}$LEP^{AA}, відповідно. Проведено генетичну паспортизацію бугаїв 25 порід за стандартними мікросателітними панелями (BM1824, BM2113, INRA023, SPS115, TGLA122, TGLA126, TGLA227, ETH10, ETH225, ETH3) для обгрунтування доцільності тривалого зберігання біологічного матеріалу від них у Національному банку генетичних ресурсів. **Висновки.** Комплексний моніторинг племінних ресурсів великої рогатої худоби за ДНК-маркерами створює підгрунтя для впровадження в Україні генетичної експертизи походження племінних тварин відповідно до міжнародних рекомендацій ISAG, що дасть змогу визначати специфіку генофонду породи чи виду загалом, характеризувати спрямованість мікроеволюційних процесів у популяціях тварин та вирішувати інші селекційно-генетичні проблеми.

Ключові слова: велика рогата худоба, поліморфізм ДНК, локуси кількісних ознак (QTL), генотип, мікросателіти (STR).

Использование молекулярно-генетических маркеров в селекционном процессе животноводства Украины

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Цель. Анализ генетической структуры быков голштинской и симментальской пород различных племпредприятий Украины по полиморфизму генов каппа-казеина, β-лактоглобулина, гормона роста, лептина, гипофизарно-специфического фактора транскрипции, миостатина. **Методы.** Индивидуальные генотипы животных идентифицировали с помощью полимеразно-цепной реакции (ПЦР) с последующим определением полиморфизма длин рестрикционных фрагментов (ПДРФ-анализ). **Результаты.** Установлен комплексный модельный генотип для повышения удоя животных украинской черно-пестрой молочной породы – к-Cn^{AB}βLG$^{AB}GH^{LV}$Pit-1^{AA} LEP^{AA}, жирномолочности – к-Cn^{AB}βLG$^{AB}GH^{VV}$Pit-1^{AA} LEP$^{BB/AB}$; украинской красно-пестрой молочной породы – к-Cn^{AA}βLG$^{AA}GH^{LL}$PIt-1^{AB}, и к-Cn^{AA}βLG$^{AB}GH^{LV}$ Pit-1^{AB}; симментальской породы – к-Cn^{BB}βLG$^{BB}GH^{LL}$ LEP^{AB} и к-Cn^{BB}βLG$^{BB}GH^{LV}$LEP^{AA}, соответственно. Проведена генетическая паспортизация быков 25 пород по стандартным микросателлитным панелям (BM1824, BM2113, INRA023, SPS115, TGLA122, TGLA126, TGLA227, ETH10, ETH225, ETH3) для обоснования целесообразности длительного хранения биологического материала от них в Национальном Банке генетических ресурсов. **Выводы.** Комплексный мониторинг племенных ресурсов крупного рогатого скота по ДНК-маркерам создает почву для внедрения в Украине генетической экспертизы происхождения племенных животных в соответствии с международными рекомендациями ISAG, что позволит определять специфику генофонда породы или вида в целом, характеризовать направленность микроэволюционных процессов в популяциях животных и решать другие селекционно-генетические проблемы.

Ключевые слова: крупный рогатый скот, полиморфизм ДНК, локусы количественных признаков (QTL), генотип, микросателлиты (STR).

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