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Allelic Polymorphism of the Somatotropin Gene among Holstein-Friesian Breed in the Livesock Farming in the Bryansk Region

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Abstract: The allelic SOMATOTROPIN gene incidence was studied among the sample of Holstein-Friesian cows at the Snezhka-Gomosoma in the Bryansk Region by versions of AluI and MspI markers and relations between different alleles and milking productivity. The study of relation between the animal genotype by the SOMATOTROPIN gene using the AluI- and MspI – markers and the milking productivity has revealed that the best productive animals are those possessing the complex genotype LL (-/-) ($p \leq 0.05$). The alleles (+)- noticeably reduce the milk yield with the stable milking registered in the animal group with the genotype LV (+/+).

Key words: Cattle · SOMATOTROPIN growth hormone · Allelic polymorphism and diary productivity

INTRODUCTION

SOMATOTROPIN growth hormone (GH) is the most essential controller of animal growth; it participates in the formation of milk productivity of cows; it is known to be linked with certain allelic genes and protein and fat concentration in milk [1-3]. The cow gene GH is located in the 19-th chromosome [4]; it includes 5 exons and 4 introns [5].

Cows have four SOMATOTROPIN alleles [6], the availability of alanine or phenylalanine at the N-end or leicin or valine in position 127 characterizes these variants. The polymorphism of the somatotrpin N-end results from the mismatch of the signal peptide reading [7]. The variants in polypeptide position 127 results from the pinpoint mutation (CTG?GTG) in the nucleotide series [8].

Two more alleles Msp(+) and Msp(-) result from the mutation localized in the third somatotropin gene introne in position 1547 resulting in site restriction for the endonuclease MspI [9]. The somatotropin alleles can be informative markers when rating the animal milking potential at early age [10]. The aim of the present work was the exploration of incidence of allelic variant of the somatotropin gene by AluI and MspI markers and their relation to the milk productivity of the Holstein-Friesian sample in the Bryansk Region.

Materials and methods. The allelic polymorphism of somatotropin genes has been studied among the Holstein-Friesian breed cows (n=55) at the Snezhka-Gosom dairy farms in the Bryansk Region. The DNA was separated according the earlier described method [11].

The polymorphism of the somatotropin gene was determined by the MspI-marker (GH/MspI) with the method of restricted length polymorphism synthesizing the DNA fragment 329 paired-end tags using primers [12]:

1) GH/MspI: 5'- CCC-ACG-GGC-AAG-AAT-GAG-GC-3'
2) GH/MspI: 5'- TGA-GGA-ACT-GCA-GGG-GCC-CA -3'

If the DNA disintegration with MspI retitase yields fragments 224 n—paired-end tag and 105 n-paired-end tags, it is the gene (+)-allele, in the opposite case; the gene has (-)-allele.

When registering the polymorphism by AluI-marker, the DNA was amplified with 223 paired-end tags using primers [13]:

1) GH/AluI: 5'- GCT-GCT-CCT-GAG-GGC-CCT-TCG-3'
2) GH/AluI: 5'- GCG-GCG-GCA-CTT-CAT-GAC-CCT -3'

The DNA restriction with the enzyme AluI results in appearance of 171 paired-end tags and 52 paired-end tags; it is the gene L-allele; the absence of disintegration indicates the V-allele.

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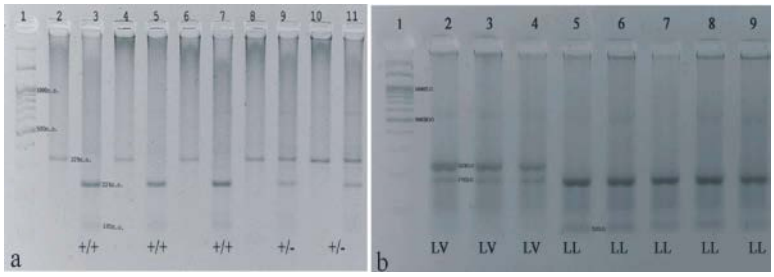

Fig. 1: Electrophoresis analysis of products of restriction of fragments of gene GH: a – GH by marker MspI, b – GH by Alu-marker: trek 1 – marker of molecular masses of M27 (SibEnzme). Characters indicate genotypes of specimens.

The fat content in milk was determined with the standard Herber method, the protein was synthesized with the GH/MspI using coloring agent Amido black 10B [14].

Main part. The work explored the allelic polymorphism of two areas of the cattle growth hormone - introne III and exon V using the restricting endonuclease MspI and AluI, respectively.

When determining the somatotropin genotype with MspI-marker using the DNA samples the fragment were amplified 329 paired-end tags long. After treatment with MspI retitase, the animal genotypes were registered: to the genotype (+/+) corresponded fragments 224 and 105 paired-end tags, to the genotype (+/-) – 329, 224 and 105 paired-end tags corresponded to the genotype (-/-) – 329 paired-end tags. Figure 1a shows the electrophoregram of products of restriction of the fragment of the somatotropin gene MspI.

After treatment of the polymorphism chain with 223 paired-end tags using the endonuclease AluI showed the growth hormone alleles L and V by the AluI-marker (Figure 1b).

The results show that the incidence of allele (+) exceeds considerably the incidence of allele (-). The cows with the genotype (+/+) are come across more often resulting in domination at the genotype (+/+) against the background of rare incipience of animals with the genotype (-/-).

To reveal the involvement of different somatotropin alleles by AluI- and MspI-markers in milk yield of cows, the correlation between gene alleles and the milk yield parameters was explored; Table 1 shows the results.

It makes apparent that the cows with the genotype LL have a higher milk yield and protein content in milk than the animals with the heterozygous genotype.

In the animal group with the genotype LL dominant the cows with milk yield 500-550 kg per month; there are cows yielding over 550 kg. The groups of cows with the genotype LV revealed no high milk yielding cows; over 70% yield less than 500 kg per month. The animal group with the genotype VV were absent in the group in question.

The animals with the heterozygote LV have the highest ft content in milk; it correlates with the results f other studies [15, 16].

Cows with the genotype LL manifested the highest protein percent in milk.

The cow with the genotype (-\-) by the MspI-marker has the highest milk yield; it correlates with the published data [17], the cows with the genotype (-\-) excel validly the cows with other variants of genotype by the protein content in milk. Over half of the cows with the genotype (+/+) yield less than 500 kg of milk. The group with the genotype (+/-) has 80% of cows with the milk yield 500-550 kg per month. The group with the homozygote by MspI(-)-allele have cows with the maximum milk yield.

Table 1: The milk yield of the Holstein-Friesian cows with different somatotropin genotypes by the AluI-marker and MspI-marker

Genotype	n	Mean milk yield per month, kg	Check milking, kg	Fat content,%	protein,%
LL	15	508,0±9,1*	16,82±0,95	3,6693±,0012*	4,64±0,47*
LV	6	474,7±8,7*	14,92±0,67	3,7283±0,0034*	3,67±0,56*
VV	0	-	-	-	-
+\+	19	477,3±4,4	15,80±0,47	3,694±0,009	4,104±0,017
+\-	8	506,2±5,0	16,93±0,61	3,666±0,005	4,04±0,090
-\-	6	526,6±7,1	17,30±0,21	3,60±0,002	4,765±0,029

*$p \leq 0.05$

Table 2: Milk yield of Holstein-Friesian cows with different genotypes by AluI- and MspI-markers

Combined somatotropin genotype by AluI- and MspI-markers	Mean monthly milk yield, kg
LL (-/-)	536,15±5.12
LL (-/+)	506,92±4.73
LL (+/+)	505,68±4,91
LV (+/-)	478,50±4,80
LV (+/+)	474,75±4,88

Table 2 shows the indicators of mean milk yield per month in the groups of cows identified with combined genotypes by the AluI- and MspI-markers. The table shows clearly that the cows with genotypes LL (-/-) dominates over other combinations of genotypes by the markers in question of the mean monthly milk yield.

Conclusions. The obtained results prove the dependence of milk yield parameters on the Somatotropin gene allele. The analysis of incidence of different somatotropin gene alleles by AluI– and MspI – markers has revealed that in the cow group in question dominate the (L)-allele and homozygote genotype LL, as well as the (+)-allele and homozygote genotype (+/+). The Holstein-Friesian cows with the somatotropin genotype (-\-) by MspI-marker show validly the highest milk yield ($p \leq 0.05$) per month and the protein content in milk; the least milk yield per is demonstrated by the cow group with the genotype (+\+)-. The cows with the somatotropin genotype LL by AluI-marker excel validly by milk yield and protein content in milk those with the heterozygote genotype, the latter have the highest fat content in milk.

The study of the comprehensive effect of the somatotropin genotype by AluI- and MspI – markers on three milk yield has revealed that the highest milk yield show the with the complex genotype LL (-/-) ($p=0.05$). The appearance of (+)-allele reduces this indicator considerably. The least milk yield is registered in the cow group with the genotype LV (+/+).

Conclusion. In order to promote the milk yield by dairy cattle in the Bryansk Region, it is advisable to carry out the livestock breeding to achieve higher incidence of somatotropin gene (-)-allele, which is associated with the highest, milk yield and protein content in milk.

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