GENOTYPES OF FIVE BLOOD PROTEINS' POLYMORPHISM IN VARIOUS PRODUCTION AGE OF DAIRY COWS

GENOTYPIZÁCIA BIELKOVÍN HEMOLYZÁTU A KRVNEJ PLAZMY DOJNÍC V RÔZNOM ÚŽITKOVOM VEKU

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SÚHRN

U kráv slovenského strakatého plemena odobraté vzorky krvi boli spracované elektroforetickou laboratórnou metódou na škrobovom géle za účelom identifikácie biochemických polymorfných znakov. Bol stanovený polymorfizmus v lokusoch hemoglobín (AA+AB+BB), amyláza (AA+AB+BB), transferín (AA+AD_1+AD_2+AE+D_1D_1+D_1E), albumín (AA+AB+BB) a postalbumín (AA+AB+AC+BB+BC+CC). Hodnotený súbor bol rozdelený do piatich vekových kategóriách hodnotených zvierat. Prvú kategóriu tvorili dojnice vo veku 25-48 mesiacov, druhú 49-72 mesiacov, tretiu 73-96 mesiacov, štvrtú 97-120 mesiacov a poslednú piatu nad 121 mesiacov.

Z výsledkov percentuálneho zastúpenia heterozygotných AB genotypov vyplýva, že bol pozorovaný pokles tohoto genotypu v polymorfizme hemoglobín z pôvodných 46,8 % (25-48 mesiac) na 11,1 % v kategórii dojníc nad 121 mesiacov. Heterozygotné genotypové kombinácie (suma AD_1+AD_2+AE+D_1E genotypov) v transferínovom lokuse vzostupne kulminovali z pôvodných 14,2 % v 25 mesiaci na 38,9 % v 121. mesiaci. Súčasne v polymorfizme postalbumín bol vo všetkých vzorkách krvi stanovený 100 % podiel heterozygotných AB+AC+BC genotypov v 121. mesiaci úžitkového veku kráv. Pre polymorfný systém amyláza heterozygotný AB genotyp kolísal v intervale medzi 15,4-23,7 %-ami bez výraznejšej tendencie vzostupu alebo poklesu. Heterozygotný AB genotyp v albumínovom lokuse bol vypočítaný v prvej vekovej kategórii 52,9 %, v druhej až piatej kategórii dosiahol rozmedzie 71,8-75,0 %. Z testovania homozygotných a heterozygotných genotypov polymorfných bielkovín krvnej plazmy a hemolyzátu bol vypočítaný preukazný rozdiel v albumínovom lokuse $2{,}18^{+}$ ($^{+}P \leq 0{,}05$).

KĽÚČOVÉ SLOVÁ: slovenské strakaté plemeno, genetický polymorfizmus bielkovín krvi, úžitkový vek dojníc

ABSTRACT

In this work were analysed samples of blood from 314 dairy cows Slovak spotted breed by the method of starch gel electrophoresis. The aim was to identify the biochemical polymorphic markers in blood plasma and in extract of erythrocytes. The polymorphism of hemoglobin (AA+AB+BB), amylasa (AA+AB+BB), transferrin (AA+AD_1+AD_2 +AE+D_1D_1+D_1E), albumin (AA+AB+BB) and postalbumin (AA+AB+AC+BB+BC+CC) were proved. File of dairy cows was divided to five groups by age. The first group create of dairy cows in age 25-48 months, second 49-72 months, third 73-96 months, fourth 97-120 months and the last over 121 months.

According to evaluation of heterozygotes genotypes were detect decrease into of AB genotype in polymorphism of hemoglobin from primary 46.8 % (25-48 month) to 11.1 % in the last age group of dairy cows up 121-th months. Heterozygotic combinations in transferrin locus (AD_1+AD_2+AE+D_1E genotypes) culminated from the beginning 14.2 % to 38.9 % at the first group and last one. The postalbumin polymorphism had the high rate of heterozygotes because of AB+AC+BC genotypes. Rate of heterozygous genotypes AB in the polymorphism system amylasa variance between 15.4 - 23.7 %. Heterozygotic genotypes AB in albumin locus was according to our analyse at dairy cows in the first evaluated of age group 52.9 %, second one only in fifth groups at intervals 71.8 - 75.0 %.

Among tested heterozygotes (AB genotype) and homozygotes (AA+BB genotypes) we found the significant differences only in albumin locus $2{,}18^{+}$ ($^{+}P \leq 0{,}05$).

KEY WORDS: Slovak spotted breed, genetic polymorphism of proteins in blood, production age of dairy cows

INTRODUCTION

The positive selection effects between genetic variants of polymorphic proteins and phenotypic characteristic of domestic animals have been published [4, 5, 6, 7, 8, 10, 12, 14, 19, 20, 21, 22].

Theoretically it is based on pleiotropic effect of genes, as alleles determining polymorphic character as in processes influencing manifestation of utilitarian characteristics [17], which are considerably modified for certain part of population and time period. Utilization of genetic polymorphic characters with heterozygotic allele organisation is stated in many experimental works. The highest effect of heterozygosis is expected when dominant and recessive pairs of alleles (AA, bb) are equally distributed with heterozygotic ones (Cc, Dd ect.).

In the characters and characteristics with low coefficients of heritability (h^2<0.30), which are considerably influenced by non-additive effects of genes, effects of heterosis is demonstrated after crossing of domestic animals. The effect is approved by higher vitality, enhanced tolerance to unfavourable environmental conditions, better fertility and growth characteristics and consequently in advanced production [2, 3, 7, 9, 15, 18].

Breeding of selected cattle population aimed on increasing of heterozygous genotype combinations for polymorphism of transferrin, celuloplasmin and amylase systems exhausted in better reproduction and body development results [13]. Kuciel and Marek [11] in study of blood serum transferrins of Czech spotted bulls indicates the highest percentage of fertility (54.74 %) after all inseminations when individuals with various heterozygotic genotypes for transferrines were used. Calculated variance confirms convenience of cattle selection with heterozygous combinations in transferrin locus [Dvořák, 1985, quotation from Bardún, 1].

The aim of the study was determine genetic composition of polymorphic proteins of blood plasma and extract of erythrocytes of Slovak spotted cows between 25-121 months of age by method of gel electrophoresis.

MATERIAL AND METHODS

Totally 878 standardised lactations of Slovak spotted dairy cows were evaluated, with minimal 87.5 % portion of the breed.

Polymorphic proteins from blood plasma and blood hemolysate were analysed by the method of starch gel electrophoresis using direct current electric source Multidrive XL. Protein fractions in polymorphism hemoglobin, transferrin, albumin and postalbumin were identified by using amidoblack 10 B solution and for determination of amylase 40% ethanol was used.

Parameters of polymorphic proteins were analysed using the Statistical Analysis System version 8.2 [16]. Heterozygotic and homozygotic genotype combinations were mutually compared by Student t-test in following five groups of age. The first group create of dairy cows in age 25-48 months, second 49-72 months, third 73-96 months, fourth 97-120 months and the last over 121 months.

RESULTS AND DISCUSSION

Homozygous genotypes combinations were identified in each polymorphic systems – hemoglobin (AA+BB), amylase (AA+BB), transferrin (AA+D_1D_1), albumin (AA+BB) and postalbumin (AA+BB+CC). Heterozygous AB genotypes were analysed in polymorphism of haemoglobin, amylase and postalbumin. Table 1 shows that heterozygous genotypes were determined in transferrin locus and genotypes in postalbumin polymorphism.

Kuciel and Marek , [11] states in samples of blood serum of Czech spotted cattle (n=200) additional four genotypes for transferrin locus D_2D_2, EE, D_1D_2 a D_2E. In contrast with our findings, Řehout and Dvořák and Čítek [14] found monomorphic AA genotypes for hemoglobin in Czech red cattle breed. In this study are stated also

Tab. 1: Genetic structure of polymorphism of proteins in blood plasma and in extract of erytrocytes dairy cows Slovak spotted breed

Polymorphic of protein	Genotypes	
	heterozygotes	homozygotes
hemoglobin (Hb)	*AB*	*AA, BB*
amylasa (Am)	*AB*	*AA, BB*
transferrin (Tf)	*AD_1, AD_2, AE, D_1E*	*AA, D_1D_1*
albumin (Al)	*AB*	*AA, BB*
postalbumin (Pta)	*AB, AC, BC*	*AA, BB, CC*

Tab. 2: Portition of dairy cows Slovak spotted breed by production of age and genotypes combination of polymorphism proteins in blood

Polymorphic protein	Genotypes	Age in month					Count (n)	Frequency (%)	Differences Student t-test
		25-48	49-72	73-96	97-120	>121			
Hb	AA	101	230	136	62	16	545	62.1	AB : (AA+BB) 0.39 -
	AB	89	155	62	10	2	318	36.2	
	BB	-	5	10	-	-	15	1.7	
Portion of heterozygotes genotypes		46.8	39.7	29.8	13.9	11.1	318	36.2	
Am	AA	129	314	145	55	14	657	74.8	AB : (AA+BB) 1.30 -
	AB	45	60	42	14	4	165	18.8	
	BB	16	16	21	3	-	56	6.4	
Portion of heterozygotes genotypes		23.7	15.4	20.2	19.4	22.2	165	18.8	
Tf	AA	-	7	-	3	-	10	1.1	$(AD_1+AD_2+AE+D_1E)$: $(AA+D_1D_1)$ 1.86 -
	AD_1	20	49	41	25	7	162	18.1	
	AD_2	3	17	8	-	-	28	3.1	
	AE	2	-	5	-	-	7	0.8	
	D_1D_1	163	306	136	44	11	660	73.8	
	D_1E	2	8	18	-	-	28	3,1	
Portion of heterozygotes genotypes		14.2	19.7	34.6	34.7	38.9	225	25.1	
Al	AA	2	38	31	7	4	96	14.4	AB : (AA+BB) 2.18 +
	AB	99	203	130	46	18	470	70.6	
	BB	86	44	38	7	2	100	15.0	
Portion of heterozygotes genotypes		52.9	71.8	65.3	76.7	75.0	196	29.4	
Pta	AA	-	13	4	-	-	17	1.9	(AB+AC+BC) : (AA+BB+CC) 0.62 -
	AB	99	179	121	46	14	459	52.3	
	AC	4	15	20	3	4	46	5.2	
	BB	57	106	40	13	-	216	24.6	
	BC	30	76	23	10	-	139	15.8	
	CC	-	1	-	-	-	1	0.2	
Portion of heterozygotes genotypes		70.0	69.2	78.9	81.9	100.0	234	26.7	

-P≥0,05 +P≤0,05

BC and CC genotype combinations in amylase locus, not identified in our blood samples.

Together with genetic analyse of proteins we have focused also on quantification of heterozygotic genotype combinations (Tab. 2). Methods used is pertinent to previous articles [10, 19, 20, 22], where we done determination of heterozygotic and homozygotic genotypes coding milk of proteins depending on lactation and age of Slovak spotted dairy cows.

Rate of heterozygotic genotypes in hemoglobin locus for first group of age (25-48 month) was 46.8 %. Following production periods of Slovak spotted cows showed tendency of decreasing incidence of genotypes AB from initially percentage 39.7 % (in 49-72 months) to 11.11 % (in age over 121 months).

While in the age of 25-48 months heterozygous AB genotype amylase represented 23.7 %, in following four groups was for this genotype observed slight increase, respectively decrease of AB genotype within interval 15.4–22.2 %.

Heterogeneity of transferrin cattle genotypes was studied as well. Most distinct difference was counted between first and fifth group of Slovak spotted cows, when heterozygous genotypes (AD_1+AD_2+AE+D_1E genotypes) increased from 14.2 % to 38.9 %.

In albumin locus heterozygous AB genotype reached on beginning of the testing period 52.9 %, while in following four groups of age was observed slight increase of this marker at intervals 71.8-75.0 %.

Polymorphism of albumin showed definite increase of heterozygous genotypes (AB+AC+BC) from initial 70% in first group of age to final 100 % in cows 121 months old.

Compendious compare of heterozygotic (AB) and homozygotic (AA+BB) genotype combinations shows statistically significant difference in biochemical polymorphism for albumin 2.18^{+} (Table 2).

CONCLUSION

Using electrophoretic method on starch gel, following data on genetic research of proteins' polymorphism in blood were obtained:

1. Genotype structure for polymorphism of hemoglobin, amylasa, transferrin, albumin and postalbumin systems.

2. Heterozygous and homozygous composition of alleles of genetic polymorphic characters in dairy cows of Slovak spotted breed within age of 24 - 121 months.

3. Testing of averages between heterozygous and homozygous genotype combinations for albumin locus showed statistically significant difference in polymorphism (t=2.18).

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