# Genetic Variants of Milk Proteins and Indexes of Milk Performance Dairy Cows of Slovak Spotted Breed on Different Lactations

A. Kúbek, J. Žitný, J. Bujko and A. Trakovická Deparment of Animal Genetics and Breeding Biology, The Slovak Agricultural University in Nitra, Slovak Republic

Abstract: The genetic structure of polymorphic milk proteins in alpha<sub>s1</sub>-casein, beta-lactoglobulin, beta-casein and kapa-casein systems were analysed by the method of starch gel electrophoresis. The homozygotic and heterozygotic genotype combinations of dairy cows of Slovak spotted breed (n = 345) were evaluated for dairy parameters. Data used in protein index calculation were obtained from lactation records of 1250 standardized lactations. 1. In polymorphism alpha s <sub>1</sub>-casein with genetic variant BC highly significance has occurred ( $^{++}P \le 0.01$ ) in milk production on the first lactations 719 kg (BC>BB) and in beta-lactoglobulin system within second and following lactations 150.6 kg of milk (AB>AA+BB,  $^{+}P \le 0.01$ ). 2. Protein content (kg) tested between groups of heterozygote and homozygote genotypes of polymorphic milk protein was significantly higher ( $^{+}P \le 0.01$ ) by 5.4 kg on second and following lactations in group of AB genotypes of beta-lactoglobulin (AB>AA+BB). 3. Within first lactations significantly high difference of 28 kg of fat has occurred between genotypes BC and BB of alpha <sub>s1</sub>-casein ( $^{++}P \le 0.01$ , BC>BB). The work is reporting also index of protein production , which is on the first lactations 16.7, on second and following lactations 105.9 and over all lactations 107%.

Key words: Dairy cows, Slovak spotted breed, polymorphic proteins, milk performance, proteins production index

#### Introduction

The input of breeding works in cattle breding is aimed to increase of milk production, which is rated as the most important producting indicator. Ability of milk synthesis and excretion is however not equal among individuals of the same breed, because various internal and external factors are taking part in this process. From these factors are essential age, gravidity, climatic factors, nutrition, tratment and milking technology, length of non-milking period, length of service-period, month of calve, movement etc.

The most of publications dealing with milk cattle breeding is giving emphasis on animal genetic foundation (Bujko, 1997; Kadleèík et al., 1992; Nový et al., 1988; Trakovická et al., 2002a; Žitný et al., 1996; Žitný et al., 2002a and Žitný et al., 2002b).

Utilize of milk protein genotyping analyses enables breeders to exert in their breeding work individuals with optimal genetic variant, hereby in short time affect occurrence of desirable alel in population (Chrenek *et al.*, 1996). Animal genotype share on producting characteristics moves from 10 to 20%. Genetic trend in west countries is characterised by improving 30-50 kg of milk and 2-3 kg of proteins per year (Hreben *et al.*, 2001). Yearly genetic advancement in particular milking cattle breeds in the world is reaching 30 – 180 kg of milk (Pribyl and Huba, 1997), in our breeds and producting types is between 36 – 97 kg of milk per

year (Bulla, 1995 and Bujko et al., 2001).

Recognition of particular effects contribution taking part in breeding work is crucial. The effect of heterosis is most impor tant one in case of non-additive gene action (Gava-lier, 1983a and Gavalier, 1983b). Genetical relativity of heterosis is coupled with raising level of heterosis in selected criteria, compared with steady state of animal population kept under equal conditions (Kúbek *et al.*, 1997; Trakovická *et al.*, 1997; Trakovická *et al.*, 2002b and Žitný *et al.*, 2002).

According to Nový et al., 1981, the highest positive effect of heterosis is assumpted when dominant and recesive pairs of alels (AA, bb) are equally distributed within heterozygot sets (Cc, Dd etc.). The premise is based on cognition, that optimal genotyp for achieving heterosis is that which cover adequate part of dominant endowments and favourable portion of homozygote and heterozygote locuses.

The current status of knowledges in the genetic of quantitative attributes shows, that heterosis is manifested mainly in attributes with low heritability coefficient (milk production). When crosses are mated inter-se, or back-crossing is used, heterosis effect within consecutive generations is declining because of gene recombining—so called recombining loss (Gavalier et al., 2000). Utilitarian characteristics with medium or high heritability coefficient (feeding and carcass characteristics), shows on the contrary low or zero

effect of heritability.

The aim of this study is to compare content of basic milk component within Slovak spotted breed dairy cows on various lactations, using genetic structure of homozygote and heterozygote locuses according to polymorphism of milk proteins.

# **Materials and Methods**

From the list of protected herds of Slovak spotted breed in the year 2002 (http://web.stonline.sk/zchssdlv/) we have taken cows (n = 345) from agri-coop Orechová Potôò, where parameters of milk production for 1250 standardized lactations were compared and statistically evaluated. Milk samples were processed using starch gel electrophoresis direct current used Multidrive XL in alkaline borate puffer pH of 8,7. On starch jellies four variants of polymorph proteins were analysed: alfa,1casein, beta-laktoglobulin, beta-casein a kapa-kasein. All selected parameters of milk production were evaluated on first, second and following by means of PC programmes.

Differencies in production of milk (kg), proteins (kg), fat (kg) between tested groups of homozygotes and heterozygotes genotypic combinations of polymorph milk proteins were tested by Student t-test on three significance levels \*P≤0.05, ++P≤0.01, +++P≤0.001. Index of protein production according to Gavalier, 1995; Psota et al., 1998, was also calculated. This index shows percentage portion of actual protein production to the limit 132 kg on 1st lactation, respectivelly 165 kg on 2<sup>nd</sup> and following lactations corrected to 3,3 % protein contect in milk. Actual protein production for each dairy cow was calculated separately (kg, %), on 2nd and following lactations as an weighted mean (Baky-tová et al., 1979). Medium values of weighted means for evaluated parameters - arithmetical means - were assigned for protein production in kilograms and protein production in percentage and appointed to the following formula:

GIBM= 
$$\left[\frac{\text{production proteins in kg}}{132 \text{ resp. } 165} + \frac{\text{production proteins in } \% -3,3}{3}\right].100$$

First calved cows on its first lactations taken into calculation were selected using following criteria:

Indexes	Limit selekcion of dairy cows				
	on 1 <sup>st</sup> lactation				
Milk (kg)	1500- 9000				
Proteins (kg)	35-360				
Proteins (%)	2,5-4				
Fat (kg)	45- 455				
Fat (%)	3 - 5,5				

Milk productions of cows on second and following lactations were selected using following parameters:

Indexes	Limit selekcion of dairy cows
Milk (kg)	2000 - 9000
Proteins (kg)	50 - 360
Proteins (%)	2,5 - 4
Fat (kg)	60 - 495
Fat (%)	3 - 5,5

### **Results and Discussion**

Genetic polymorphism of milk proteins of dairy cows of Slovak spotted breed on the first lactation is, that in polymorphism of alpha<sub>s1</sub>-casein the cows with heterozygotic BC genotype has statistically high significant difference of 719 kg milk (BC>BB,  $^{++}P \le 0.01$ ).

Milk production on second and subsequent lactations (Table 1) was increasing in the group of heterozygotes as follows: kapa-Cn (BC > AA + BB, difference 52,6 kg of milk), alpha, -Cn (BC>BB, difference 135,9 kg of milk), beta-Cn (AB + AC + BC > AA + BB + CC, difference 40,1 kg of milk). Just in polymorphism of beta-Lg statistically significant difference between heterozygotic and homozygotic genotypes was detected 150,6 kg of milk (AB > AA + BB,  $^+P \le 0.01$ ). From the view of milk production are differences for all lactations and within all four polymorphic protein systems non-significant. In the 1st group of heterozygote genotypes, cumulating AB+AC+BC capa-casein genotypes, the highest milk production of 4751,5 kg per lactation was recorded. žitný et al., 1997 cited, that in summary evaluation of performance characteristic of the Slovak spotted breed (n = 717) using genetic variants of alpha<sub>s1</sub>-casein on first four lactations, statistically significant differences (P = 0.05) were found in milk (BC>BB) and lactose (BC>BB) production.

Differencies in protein production in kg by lactations are summarised in Table 2. Statistic differences between homozygotic and heterozygotic genotype combinations on first lactation are statisticaly non-significant within interval 1,0 – 14,8 kg of proteins.

The highest average protein production 175,4 kg was achieved on second and following lactations in the group of AB beta-lactoglobulin genotypes, with significant difference of 5,4 kg of proteins  $(AB>AA+BB, ^+P\leq 0.01)$ .

Differences of protein production markers (kg) for all lactations toghether were rising in the group of heterozygotic genotypes in this order: in polymorphism kapa-casein AB genotyp (2.0 kg of proteins), alpha s1-casein BC genotyp (2.1 kg of proteins), beta-casein AB + AC + BC genotypes (2.6 kg of proteins) and finaly

Table 1: Statistical characteristic of milk production (kg) in cows of Slovak spotted breed according to

Lactation	1 <sup>st</sup> group Heterozy	gotes gen	otypes		_	2 <sup>nd</sup> group Homozygotes genotypes				
	Count lactation	×	s <sub>x</sub>	v	Count lactatio	x on	S <sub>x</sub>	v	t-test 1 : 2	
	α, - casei	n								
First lactation	14	4689,7	897	19	325	3970,7	985	24	719,0++	
Second and	26	5072,0	1118	22	885	4936,1	1101	22	135,9 <sup>-</sup>	
Following lactat	tion									
All lactation	40	4938,2	1051	21	1210	4676,8	1153	24	261,4 <sup>-</sup>	
	β-lactogl									
First lactation	155	3982,0	984	24	184	4016,0	999	24	-34,0	
Second and	419	5021,3	1123	22	492	4870,7	1078	22	150,6 <sup>+</sup>	
Following lactat	tion									
All lactation	574	4740,7	1181	24	676	4638,1	1123	24	102,6	
•	β-casein									
First lactation	153	4054,6	959	23	186	3955,9	1017	25	98,7 <sup>-</sup>	
Second and	399	5018,7	1037	20	512	4878,6	1146	23	140,1	
Following lactat	tion									
All lactation	552	4751,5	1103	23	698	4632,7	1185	25	118,8 <sup>-</sup>	
	K-casein									
First lactation	205	4038,9	991	24	134	3941,6	992	25	97,3 <sup>-</sup>	
Second and	651	4960,2	1092	22	350	4907,6	1116	22	52,6 <sup>-</sup>	
Following lactar	tion									
All lactation	766	4713,6	1141	24	484	4640,2	1165	25	73,4	

 $^{+}P \ge 0.05 \quad ^{+}P \le 0.05 \quad ^{+}^{+}P \le 0.01 \quad ^{+}^{+}^{+}P \le 0.001$ 

Table 2: Statistical characteristic in production of proteins (kg) in cows of Slovak spotted breed according to

Lactation	ypical com 1 <sup>st</sup> grou Heteroz			ı	2 <sup>nd</sup> group Homozyg	Differences Student					
	Count lactation	X 1	s <sub>x</sub>	V	Count lactation	x	s <sub>x</sub>	v	t-test 1 : 2		
	α <sub>s1</sub> - cas	α <sub>s1</sub> - casein									
First lactation	13	159,9	33	21	272	145,1	34	23	14,8 <sup>-</sup>		
Second and	25	172,0	37	21	817	172,5	37	21	-0,5		
Following lactat	ion										
All lactation	38	167,8	36	21	1089	165,7	38	23	2,1		
	β-lactoglobulin										
First lactation	134	144,8	34	23	151	146,6	34	23	-1,8		
Second and	393	175,4	37	21	449	170,0	36	21	5,4 <sup>+</sup>		
Following lactat	ion										
All lactation	527	167,6	39	23	600	164,1	37	22	3,5		
	β-casein										
First lactation	136	146,3	32	22	149	145,3	35	24	1,0		
Second and	366	175,0	35	20	476	170,6	38	22	4,4		
Following lactat	ion			•					,		
All lactation	502	167,2	36	22	625	164,6	39	23	2,6 <sup>-</sup>		
	K-casein										
First lactation	173	146,9	32	22	112	144,0	36	25	2,9		
Second and	517	173,1	35	20	325	171,6	39	22	1,5		
Following lactat	ion	•									
All lactation	690	166,5	36	22	437	164,5	40	24	2,0		

 $P \ge 0.05 + P \le 0.05 + P \le 0.01 + P \le 0.001$ 

Table 3: Statistical characteristic in production of fat (kg) in cows of Slovak spotted breed according to

Lactation	1 <sup>st</sup> grou Heteroz	p ygotes ger	notypes	-	2 <sup>nd</sup> grou Homozy	Differences Student					
	Count lactation	x า	s <sub>x</sub>	٧	Count lactation	X	s <sub>x</sub>	v	t-test 1 : 2		
	α <sub>s1</sub> - cas	α <sub>s1</sub> - casein									
First lactation	14	205,8	38	18	324	177,8	42	24	28,0++		
Second and	26	231,8	63	27	884	227,9	54	23	3,9		
following lactati	ion								-,-		
All lactation	40	222,7	56	25	1208	214,5	55	26	8,2 <sup>-</sup>		
	β-lactoglobulin										
First lactation	155	177,5	45	25	183	180,3	40	22	-2,8 <sup>-</sup>		
Second and	418	230,2	55	24	492	226,2	53	23	4,0		
following lactati	ion					•					
All lactation	573	215,9	57	26	675	213,8	54	25	2,1		
	β-casein								,·		
First lactation	153	181,3	41	23	185	177,1	44	24	4,2		
Second and	398	230,6	50	21	512	226,0	57	25	4,6		
following lactati	ion	•					•		.,.		
All lactation	552	216,5	53	24	698	212,8	58	27	3,7 <sup>.</sup>		
	K-casein					_ , _ , _			0,,		
First lactation	204	180.2	42	23	134	177,2	43	24	3,0		
Second and	560	228,5	53	23	350	227,3	55	24	1,2 <sup>-</sup>		
following lactati		.,-				,			.,_		
All lactation	764	215,6	55	25	484	213,4	57	26	2,2		

 $P \ge 0.05$   $P \le 0.05$   $P \le 0.01$   $P \le 0.001$ 

Table 4: Statistical characteristic of index of proteins production (%)of dairy cows of Slovak spotted breed

Indexes	Literary sentences								
1 <sup>st</sup> lacta	st lactation 2 <sup>nd</sup> and following lactation All lactationxs								
x	S	V	x	s	V	x	s	٧	
-	-	-	-	-	-	97,4	26,5	27,1	Gavalier, 1995
-	-	-	89,2	16,3	18,2	90,9	17,8	19,6	Bujko, 1997
95,1	-	-	88,8	-	-	-	-	-	Psota et al., 1998
116,7	25,5	21,9	105,9	19,5	18,4	107,0	20,4	19,1	Ours results

beta-lactoglobulin with AB genotype and 3,5 kg of proteins. Not all differences are however statistically significant ( $P \ge 0.05$ ).

Concerning to another character – fat production (kg), statistically highly significant differences on first lactations were calculated just in group of heterozygotes BC genotypes of alpha  $_{\rm s1}\text{-}{\rm casein}$  28,0 kg (BC > BB,  $^{++}{\rm P} \! \leq \! 0.01$ ).

We have detected, that for milk fat production (kg) are definitely allocated differences among evaluated groups on second and following lactations, as well as on all lactations in polymorphism of heterozygote genotypes of alpha  $_{s1}$ -casein (BC), beta-lactoglobulin (AB), beta-casein (AB+AC+BC) and kapa-kasein (AB). In the Table 3 are stated testing results of all four milk proteins with statisticaly non-significant differences ( $P \ge 0.05$ ).

According to literature data, heterozygotic variants of

alpha<sub>s1</sub>-casein, beta-lactoglobulin, beta-casein and kapa-casein positively influenced all three indicators of performance dairy control – milk (kg), proteins (kg) and fat (kg). Results achievied confirms observations on other livestock animals, that organisms with higher level of heterozygotism have better total permance parameters (Nový et al., 1988; Trakovická et al., 1997 and Trako-vická et al., 2002b), thus also adaptability and resistence towards unfavourable external conditions.

Last indicator presented is the index of protein production (GIBM according to Gavalier, 1995). Average value of GIBM for first lactations is 116,7%, for second and post-lactations 105,9%, for all lactations together 107%. Following variational and statistical indicators of the protein production index (Table 4) is visible, that evaluated group of dairy cows of Slovak spotted breed is representing qualitative

superior population within protected cattle herds in Slovakia, comparing with cow sets evaluated by Gavalier, 1995; Bujko, 1997 and Psota *et al.*, 1998. We can enunciate, that production and content of proteins in milk is besides of genetic influence affected by nutrition, health status, phase of lactation and other factors of breeding environment (Chrenek *et al.*, 1996).

#### Conclusion

To achieve onwards improving performance efficiency by exploiting effect of heterosis is necessary systematicly improve average genetic values of base breeds used in crossbreeding. This cross system require disposition of thorough-bred base populations – breeds, strains and synthetic populations.

## References

- Bakytová, H., M. Ugron and O. Kontšeková, 1979. Základy štatistiky. In: Vydala ALFA, vydavateľstvo technickej a ekonomickej literatúry, n. p. Bratislava, 392 s.
- Bujko, J., 1997. Vyhodnotenie ukazovateľov produkèných a reprodukèných vlastností kráv slovenského strakatého plemena v chránenom chove Agroanim š. p. Prešov. In: Diplomová práca, SPU Nitra, 58 s.
- Bujko, J. and M. Rybanská, 2001. Genetic effect in the model herd of cows Slovak spotted breed. In: Genetics and animal breeding, 4th International PhD Student's Conference, Preroy, Czech Republic, pp. 78–80.
- Dvorák, J., Z. Havlíček and J. Pribyl, 1994. Genetical polymorphism and milk protein production. In: 45 th Annual Meeting of the EAAP Edinburg, pp. 1-24.
- Gavalier, M., 1983a. Heterosis and maternal efects in animal breeding. In: Agri. Sci., Series A, No. 3, pp: 136.
- Gavalier, M., 1983. Share of additive and heterozygotic effects on milk production and meat production at cross-breeding in cattle. In: Acta fytotech. et zootech. roè. XXXIX, 1983-b, pp: 17-23.
- Gavalier, M., 1995. Typ, exteriér a mlieková úžitkovos\_ v troch chránených chovoch Západoslovenského regiónu. In: Problémy a perspektívy rozvoja plemenárskej práce v chove hovädzieho dobytka na Slovensku. Nitra: ZCHSSD, s. 99-111.
- Gavalier, M. and M. Rybanská, 2000. Šľachtenie hospodárskych zvierat (skripta). In: Edièné stredisko SPU Nitra, 148 s.
- Hreben, F. and J. Pytloun, 2001. Lze srovnávat výpoèet plemenné hodnoty u skotu u nás a v zahranièí? In: Náš chov, è.9, s. 23–25.
- http://web.stonline.sk/zchssdlv/; Zväz chovateľov slovenského strakatého dobytka.
- Chrenek, P. and D. Vašíèek, 1996. Využitie PCR pri identifikácii ekonomicky dôležitých genetických variantov HD. In: Slovenský chov, roè. I, è.2, s. 27.
- Kadleèík, O., J. Pšenica, P. Brzuski and J. Candrák, 1992. Milk protein content and protein yield in cows of various breeds. In: Czech J. Anim. Sci., 37: 341-349.

- Kúbek, A., 1981. Polymorfné biochemické znaky vo vztahu k produkcii hovädzieho dobytka. In: Zborník vedeckej konferencie AF VŠP Nitra, s. 125-126.
- Kúbek, A., A. Trakovická, J. Itný, E. Michalièková and M. Gavalier, 1997. Stage of homozygosis of polymorphic markers and milk production of dairy cows. In: Current problems of breeding, health, growth and production of cattle. Book of abstracts of the International Conference, Èeské Budìjovice, Czech Republic, pp: 53-55.
- Mácha, J., 1991. Genetický polymorfismus bílkovin mléka a možnosti jeho využití. Folia, Fac. Agron., Rāda A, Brno, 60 s.
- Nový, J., M. Gavalier, A. Kúbek and M. Jamriška, 1981. Genetické aspekty intenzifikácie živoèíšnej výroby. In: Vydala PRÍRODA, vydavateľstvo kníh a èasopisov, n. p. Bratislava, 251 s.
- Nový, J., O. Kadleèík, J. Pšenica and J. Atkoviè, 1988. Zhodnotenie výsledkov homogenného a heterogenného priparovania. In: VŠP Nitra, Závereèná správa za subetapu P 06-529-820-03 / 05-3, 39 s.
- Psota, T., M. Gavalier, J. Uhlár, M.Kubaèka and M. Ivánek, 1998. Aký je hovädzí dobytok slovenského strakatého plemena? In: Slovenský chov, roè. III, è. 3, s. 22.
- Trakovická, A., A. Kúbek, J. Itný and S. Smatanová, 1997. The genetic polymorphism of blood and wool production of sheep. In: Acta zootechnica, Slovaca Universitas Agriculturae Nitriae, Slovakia, 52: 59-66.
- Trakovická, A., A. Kúbek and J. žitný, 2002a. The effects of CSN3 gene expresion to the parameters milk production of cows. In: Cattle in new millennium, Book of abstracts of international scientific conference, Nitra, Slovakia, pp. 217-221.
- Trakovická, A., K. Bežová and J. Benková, 2002b. Genetic polymorprism egg and blood proteins at relations to reproduction in hens. In: Poultry and small farm animal breeding in the third millenium, Book of abstracts of international scientific conference, Nitra, Slovakia, pp. 163-166.
- Žitný, J., A. Trakovická, A. Kúbek, E. Michalièková and I. Ostertág, 1996. Differences in milk efficiency of different kappa-casein genotypes of dairy cows of the Slovak pied breed. In: Czech J. Anim. Sci., 41: 533-538.
- Žitný, J., I. Ostertág, E. Michalièková, A. Kúbek, A. Trakovická and M. Gavalier, 1997. Differences in milk performance of dairy cows of the Slovakian pied breed of various á<sub>s1</sub>-casein genotypes. In: Czech J. Anim. Sci., 42: 481-486.
- Žitný, J., A. Kúbek, V. Zimmerman, A. Trakovická, K. Tothová, 2002a. Genetic analysis of milk proteins from production age aspect. In: Agriculture, 48: 389-393.
- Žitný, J., J. Bujko, A. Trakovická, E. Strapáková and K. Tothová, 2002. Evaluation of milk production of dairy cows of the Slovak spotted breed according to selection for longevity and genetic variants of polymorphic proteins in milk. In: J. Central European Agri., 3:195 204, http://www.agr.hr/jcea/.