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Full Length Research Paper

Association between milk protein polymorphism and milk production traits in Black and White dairy cattle in Turkey

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Milk protein polymorphism such as α_{s1}-casein (α_{s1}-Cn), β-lactoglobulin (β-Lg), β-casein (β-Cn) and κ-casein (κ-Cn) of Black and White dairy cattle were investigated in this study. The genetic structure of herd was examined for these loci. At the sametime the relationships between milk protein types and some milk production traits were determined. Milk production traits were chosen as milk yield, average daily milk yield and lactation length. These traits were associated with milk protein types. Genetic variants of milk protein were identified by starch gel electrophoresis containing mercaptoethanol and urea. The allele gene frequencies of A, B, and C of α_{s1}-Cn loci and A, B of β-Cn were found to be 0.01; 0.97; 0.02 and 0.95; 0.05 respectively. Furthermore, the allele frequencies of A, B of κ-Cn and A, B of β-Lg were found to be 0.68; 0.32 and 0.55; 0.45 respectively. For α_{s1}-Cn BB phenotype was found to have higher milk yield than those of α_{s1}-Cn AB and α_{s1}-Cn BC phenotypes, and β-Lg AA phenotype was detected to have higher milk yield than those of β-Lg AB and β-Lg BB types but were not statistically important. It was concluded that the genetic structure of the four loci were determined in Black and White cattle, but there was no significant association between the milk protein types and milk production traits in the study.

Key words: Milk protein polymorphism, casein, lactoglobulin, Black and White dairy cattle.

INTRODUCTION

It is the interest of animal breeders to identify the animal genotypes from the milk, blood and tissue variations with respect to such genetic variants which might be used as a tool to clarify the phylogenetic relationship. Since these traits show polymorphic structures, any association between production characteristics and genetically inherited phenotypes could be the starting point of indirect selection. Milk protein polymorphism has an advantage to follow a co-dominant inheritance and to determine the relatively small number of alleles, and also data is obtainable easily. Milk protein phenotypes also have advantage of directly reflecting genotypes of individuals due to lack of environmental effects.

Milk protein polymorphism was firstly reported by Aschaffenburg and Drewry (1957). Especially, some evidences have been found between genetic variant of milk protein and production traits for livestock animal by Hall in 1974. After that many investigations were completed by several researchers in this area. The genetic causes of this relationship between milk protein polymorphism and production traits were thought to be due to pleiotropy, linkage and heterosis (Soysal, 1983).

Milk proteins are the mixture of several of proteins (alfa-laktalbumin, beta-lactoglobulin and casein). The complex of casein is a main structure of milk protein and the ratio of casein is 2.63% in bovine milk. Bovine milk proteins are classified by two groups; one group is soluble at pH 4.6, on the other hand, the other group is not soluble at pH 4.6. The soluble part is called whey protein such as α- laktalbumin (α-La) and β- lactoglobulin (β-Lg). However, the insoluble part constitutes four casein protein such as α_{s1}-casein (α_{s1}-Cn), α_{s2}-casein (α_{s2}-Cn), β-casein (β-Cn) and κ-casein (κ-Cn) (Sekerden et al.,1993; Demirci, 1995; Ulutas and Yildirim, 2009).

Milk protein polymorphism and its relationship with economical traits in livestock animal were studied by many researchers and were argued in their papers. Some researchers reported that this relationship was not important for milk production traits (Ustdal et al., 1982; Soysal, 2000; Ulutas and Yildirim, 2009). Whereas, Mclean et al. (1985), Khaertdinov (1990), Rensburg et al. (1991) and NG-Kwai-Hang et al.(1991) recommended that genetic variant of milk protein could be a criteria of selection for the improvement of dairy cattle production.

For instance, Dayioglu and Dogru (1997) found an effect of β-Lg AA phenotypes on milk fat percentage in Brown Swiss cattle (P<0.01). Dogru and Dayioglu (1996) reported that α- Cn phenotypes on milk yield were not significant but β-Cn BB phenotypes on actual milk yield were significant (P<0.05) and κ-Cn phenotypes caused significant differences on daily average milk yield and actual fat yield (P<0.05). Cornberg et al. (1964) and Samarineanu et al. (1984) found an association of β-Lg AA phenotype with milk yield. β-Lg BB phenotype was related to higher milk and fat yields than the other phenotypes by Kriventsov and Prozorov (1975). Macha et al. (1984) reported that α-Cn BC locus had the highest ratio of milk protein than the other loci. Similarly, β-Lg AA and β-Lg BB phenotypes were found as higher milk yield than β-Lg AB for Black and White cattle breed by Mayer et al. (1990) (P<0.05). Conversely, Wegner et al. (1974), Janicki et al. (1980), Nakayama et al. (1996), Eidrigevich et al. (1982), Zogg (1990) and Sekerden et al. (1999) reported that milk protein polymorphism had no significant effect on milk production traits. Especially, Sekerden et al. (1993) reported that the relationship with milk protein loci and production traits such as milk yield, milk fat yield was not significant related with the α- Cn phenotype in Jersey cattle. Moreover, Ulutas and Yildirim (2009) did not report any relationship between milk yield, fat content and genotypes of β-Lg, β-Cn and αs_1-Cn (P>0.05), but rennet clotting time of milk was found to be significant related with genotype of β-Lg , and β-Cn, except αs_1-Cn.

The aim of this study was to determine the genetic structure of herd in terms of α_{s1}-casein, β-lactoglobulin, β-casein and κ-casein genotype and also to investigate some relationship between milk protein types and some production traits.

MATERIALS AND METHODS

The research material consisted of 90 Black and White dairy cattle and their milk samples. The Black and White cows were reared in Tahirova State Farm. Totally 209 production records, covering the period from 1989 to 2000, were used for the study.

Milk samples were taken into 10 ml sterilized tubes and stored at -20°C until electrophoresis in deep freeze. All milk samples were centrifuged at the rate of 1300 rpm for 20 min in order to separate the milk fat layer from milk samples and whole studies were conducted with milk samples without fat. Starch-urea gel electrophoresis was used to identify α_{s1}-Cn, β -Cn, κ-Cn and β -Lg phenotypes of the milk proteins according to Aschaffenburg and Thymann (1965). After electrophoresis, the gels were coloured with amido black 10 B for about 20 min and was discoloured according to Aschaffenburg and Machalak (1968).

Gene frequencies with standard errors were calculated by direct counting method for genetic variant of milk proteins according to Soysal (2000). The deviations observed gene frequencies of α_{s1}-Cn, β -Cn, κ-Cn and β -Lg phenotypes of the milk proteins alleles from the expected frequencies under the assumption of genetic equilibrium were investigated by chi-square test (χ^2) (Soysal, 1998).

Statistical methods

The variation of average daily milk yield, lactation length and milk yield for milk production traits were investigated by Least Square Analysis of SAS (1992). The following mathematical model was used:

$$Y_{ijklm} = \mu + l_i + YMLS_j + PF_k + b(SG_1) + e_{ijklm}$$

Where Y_{ijklm}= is the dependent variable (avarage daily milk yield, milk yield and lactation length); μ= overall population mean; l_i= randomized effect individuals; YMLSj= fixed effect of calving season, the number of lactation and production year; PFk=fixed effect of milk protein genotypes; $b(SG_1)$= the regression of lactation length on production traits; e_{ijklm}= random error.

The calving year was evaluated as production year for the 11 years of production records obtained between 1989 and 2000. Four calving season were included, such that every three months of the year starting from the last month of the previous year were con-sidered as one group of seasons as spring, summer, autumn and winter. Five groups for number of lactations were included in the model. Lactation milk yields records were adjusted according to Anonymous (1976).

RESULTS

Genetic polymorphisms of α_{s1}-Cn, β-Lg, β -Cn and κ-Cn phenotypes were studied in this work. The distribution of α_{s1}-casein (α_{s1}-Cn), β-lactoglobulin (β-Lg), β-casein (β-Cn) and κ-casein (κ-Cn) phenotypes and their percentages are showed in Table 1. In addition, gene frequencies and their standard errors for genetic variants of the four milk protein systems are given Table 2. In this study, the population was balanced in the Hardy-Weinberg equilibrium.

AA and AB phenotypes of β-Cn were observed but BB phenotype was not found in the study. The A, B and C allele frequencies of α_{s1}-Cn loci were found as 0.01, 0.97 and 0.02, respectively. The A and B allele frequencies of β-Lg loci were found as 0.55 and 0.45 respectively. Likewise, the A and B allele frequencies of κ-Cn loci were detected as 0.68 and 0.32, respectively. Also the A and B allele frequencies of β-Cn loci were determined as 0.95 and 0.05, respectively.

This study showed that κ-Cn A gene (0.68) frequency was higher than B gene (0.32) in Black and White cows. Moreover, β-Cn A gene (0.95) frequency was higher than B gene (0.05) in Black and White animal. The A and B

Table 1. The distribution of protein genotypes and their percentages (%).

Locus	α_{s1}-Cn			β-Cn			κ-Cn			β-Lg		
Genotype	BB	AB	BC	AA	AB	BB	AA	AB	BB	AA	AB	BB
Number	85	2	3	82	8	--	46	32	12	33	34	23
Percentage (%)	95	2	3	91	9	--	51	36	13	37	38	25

Table 2. The distributions of allele frequencies for protein genotypes.

Locus	β-Lg		β-Cn		κ-Cn		α_{s1}-Cn		
Allele	A	B	A	B	A	B	A	B	C
Frequency	0.55	0.45	0.95	0.05	0.68	0.32	0.01	0.97	0.02
Standard error	0.03	0.03	0.01	0.01	0.03	0.03	0.0078	0.012	0.01

Table 3. Least square means with standard error for protein genotypes and various production traits.

Locus	Genotype	Number	Average Daily Milk Yield (kg)	Milk yield (kg)	Lactation length (day)
α_{s1}-Cn	BB	200	16.74 ± 0.35	5071 ± 156.62	298 ± 5.60
	AB	4	15.82 ± 1.22	4590 ± 354.59	285 ± 6.07
	BC	5	16.70 ± 1.52	4119 ± 191.70	277 ± 27.80
β-Cn	AA	193	16.52 ± 0.36	4984 ± 157.03	296 ± 5.85
	AB	13	17.09 ± 1.57	5104 ± 319.30	287 ± 19.44
κ-Cn	AA	90	18.14 ± 0.61	5481 ± 191.03	303 ± 8.85
	AB	27	17.35 ± 0.59	5226 ± 187.30	305 ± 8.44
	BB	22	17.27 ± 1.15	5510 ± 361.09	327 ± 16.45
β-Lg	AA	92	18.22 ± 0.59	5445 ± 187.08	309 ± 8.45
	AB	68	17.19 ± 0.70	5393 ± 219.78	307 ± 10.15
	BB	49	17.10 ± 0.87	5142 ± 275.09	301 ± 12.60

allele frequencies of β-Lg loci were found as 0.55 and 0.45, respectively.

Association between α_{s1}-Cn, β-Lg, β-Cn and κ-Cn phenotypes and some production traits was investigated. Statistical results are summarized in Table 3 for the production traits according to the phenotype of α_{s1}-Cn, β-Lg, β-Cn and κ-Cn. Fixed effect of calving season, number of lactation and production yield were found as the very important factor for the milk production traits ($P < 0.01$) but genotype of milk protein was not detected to be statistically important.

In this study, the relationship between α_{s1}-Cn phenotypes and production traits was not statistically significant. Likewise, the highest milk production was obtained from α_{s1}-Cn BB phenotype. β-Lg phenotype when compared with milk production traits and β-Lg phenotypes had no relationship. Only two phenotypes of β-Cn loci were observed (AA and AB) and β-Cn AB was higher than β-Cn AA for the milk yield. The highest and lowest κ-Cn phenotypes for milk yield was obtained from κ-Cn BB and κ-Cn AB, respectively in the study but the all differences were not important statistically ($P>0.05$).

DISCUSSION

Gene frequencies were compared with some previous studies. AA and AB genotypes of β-Cn was observed but BB genotype was not found in the study. Similar results were reported by Mariani (1989) and Anne-Marie and Kristiansen (1989). In general, the distributions of gene frequencies were in accordance with other researchers (Dogru and Dayioglu, 1996; Samorineanu et al., 1984; Mariani, 1989; Quinteros et al., 1983; Anne-Marie and Kristiansen, 1989). Exceptionally, Han et al. (1985) found A allele frequency to be higher (0.94) than the others for the α_{s1}-Cn loci.

This study showed that κ-Cn A gene frequency was higher than B gene in Black and White cows. Similar observations were obtained by Mariani (1989) and Anne-

Marie and Kristiansen (1989) in Black and White cattle breed. However, Ozdemir and Dogru (2005) reported that κ-Cn B gene frequency was higher than A gene. Moreover, β-Cn A gene frequency was higher than B gene in Black and White cattle. Similar observations were found by Han et al. (1985) and Mariani (1989). The A and B allele frequencies of β-Lg loci were detected as 0.55 and 0.45, respectively. Similar results were also reported by Stasio et al. (1982).

Phenotypes of milk protein were not found to be statistically important in this study. Similarly, Wagner et al. (1974), Janicki et al (1980), Sekerden et al. (1999), Nakayama et al. (1996), Zogg (1990) ,Ustdal et al. (1982) and Eidrigevich et al. (1982) also reported that milk protein polymorphism had no significant effect on milk production traits. Ulutas and Yildirim (2009) did not find any relationship between milk yield, fat content and genotypes of β -Lg, β-Cn and αs_1-Cn. Whereas, some other studies by Mclean et al. (1985), Khaertdinov (1990), Rensburg et al. (1991) and Ng-Kwai-Hang et al. (1991) suggested that genetic variants of milk protein was related with production trait.

In this study, the associations between α_{s1}-Cn phenotypes and production traits were not significant statistically. Likewise, the highest milk production was obtained from α_{s1}-Cn BB phenotype. Identically, Dogru and Dayioglu (1996) did not report any relationship between α- Cn phenotypes and milk yield. On the other hand, Prozonov (1973) and Macha et al. (1984) reported that α-Cn BC locus was associated with milk protein percentage. Also, genotypes of αs_1-Cn were found to be significant for milk yield, milk fat yield and 305-days milk yield in Black and White cattle breed (Ozdemir and Dogru, 2004) ($P<0.05$).

There was no relationship between β-Lg phenotype and milk production traits. Cornberg et al (1964) and Samarineanu *et al*. (1984) found an association of β-Lg AA phenotype with milk yield in Black and White cattle and Brown Swiss cattle breeds, respectively. Similarly, Munro (1979) reported highly significant differences among β-Lg phenotypes for milk protein percentage. Likewise, β-Lg AA and β-Lg BB phenotypes were associated with higher milk yield than β-Lg AB for Black and White cattle breed by Mayer et al.(1990) ($P<0.05$). β-Lg AA and β-Lg AB genotypes may have selective advantage in terms of some milk production traits (Ozdemir and Dogru, 2007).

The only two phenotypes of β-Cn loci were observed (AA and AB) and β-Cn AB was found to be higher than β-Cn AA for milk yield ($P>0.05$). Dogru and Dayioglu (1996) reported that β-Cn BB phenotypes on actual milk yield were significant ($P<0.05$). The highest and lowest κ-Cn phenotypes for milk yield were obtained from κ-Cn BB and κ-Cn AB respectively in this study ($P>0.05$). Correspondingly, there were significant relationships between milk production traits and κ-Cn loci in Black and White cattle breed (Komatsu et al., 1982; Pavlyuchnko and Pubkova, 1985; Eggen and Fries, 1990; Pedersen, 1991; Dogru and Dayioglu 1996) also in Simmental cattle (Putz et al., 1992). Also, Ozdemir and Dogru (2005) suggested that κ-Cn BB and κ-Cn AB genotypes might be selective advantage for some milk production traits.

Conclusion

It was concluded that the genetic structure of herd was determined by this study in terms of α_{s1}-Cn, β-Lg, β-Cn and κ-Cn loci. No significant relationship was detected between milk yield, average daily milk yield and lactation length with genetic variant of milk protein. Thus, this relationship may be significant or not based on examined production trait. It is possible that the results are changeable for different breeds or herd within same breed according to genetic structure of these loci in the population.

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