MILK PROTEIN POLYMORPHISMS AS GENETIC MARKER IN KOREAN NATIVE CATTLE

E. R. Chung[1], S. K. Han[2] and T. J. Rhim

Department of Dairy Science, College of Agriculture
Sang Ji University, Wonju 220-702, Korea

Summary

Genetic variants of αs_1-casein, β-casein, κ-casein and β-lactoglobulin were investigated by starch urea gel electrophoresis in milk samples of 280 Korean native cattle. A new β-casein variant, designated β-casein A^4, was found in milk samples of Korean native cattle. It has a much slower electrophoretic mobility than the β-casein A^3 variant in acid gel. This new variant appeared together with either β-casein A^1, A^2 or B variant. Gene frequencies and genotypic frequencies were estimated. Gene frequencies of four milk protein loci in Korean native cattle were compared with those of imported cattle breeds raised in Korea and Japanese brown cattle. Gene frequencies were αs_1-casein B .846, αs_1-casein C .154; β-casein A^1 .216, β-casein A^2 .666, β-casein A^4 .048, β-casein B .070; κ-casein A .648, κ-casein B .352; β-lactoglobulin A .148, β-lactoglobulin B .852. The population was in Hardy-Weinberg equilibrium at all milk protein loci. Gene frequencies of Korean native cattle were very similar to those of Japanese brown cattle. Interestingly, a new variant, β-casein A^4, was found only in Korean native cattle and Japanese brown cattle. These results support the hypothesis that Korean native cattle were used in the development of the Japanese brown cattle.

(**Key Words** : Milk Proteins, Genetic Polymorphisms, Genetic Markers, Korean Native Cattle)

Introduction

Since the initial discovery of two variants of β-lactoglobulin (β-LG) in cow's milk by Aschaffenburg and Drewry (1955), genetically controlled variation of all five major milk proteins has been demonstrated (Eigel et al., 1984). Until now the αs_1-casein (αs_1-CN) locus has 5 alleles (A, B, C, D and E). β-casein (β-CN) locus shows 7 alleles (A^1, A^2, A^3, B, C, D and E). κ-casein (κ-CN) has 4 alleles (A, B, C and E) and β-lactoglobulin occurs in 9 different variants (A, B, C, D, E, F, G, H and W) and α-lactalbumin (α-LA) shows 3 alleles (A, B and C). These major milk proteins are controlled by codominant autosomal genes according to the Mendelian law of inheritance (Aschaffenburg, 1968). The various genetic variants differ from each other as a consequence of either substitution or deletion of amino acids within the polypeptide chain (Eigel et al., 1984). Charge differences in their protein molecules allow the separation and detection of the different variants by electrophoresis.

Milk protein genetic polymorphisms have received considerable research interests in recent years because of possible associations between milk protein genotypes and economically important traits in dairy cattle. Many research reports have indicated that certain milk protein variants may be associated with milk production (Ng-Kwai-Hang et al., 1984; Gonyon et al., 1987: Haenlein et al., 1987: Lin et al., 1989; Bech and Kristiansen, 1990), milk composition (McLean et al., 1984; Graml et al., 1986; Lin et al., 1986; Ng-Kwai-Hang et al., 1986; McLean, 1987; Aleandri et al., 1990; Eenennaam and Medrano, 1991; Bovenhuis et al., 1992), cheese production (Schaar et al., 1985; Marziali and Ng-Kwai-Hang, 1986; Aaltonen and Antila, 1987; McLean, 1987; Pagnacco and Caroli, 1987; Grosclaude, 1988; McLean and Schaar, 1989) and growth and reproductive performance (Hargrove et al., 1980; Singh et al., 1981; Jairam and Nair, 1983; Ronda and Perez-Beato, 1983; Lin et al., 1987). Therefore, milk protein genes might be useful as genetic markers for the additional selection criteria in dairy cattle breeding. The possibilities for genetically improving the population for a single gene are

[1] Address reprint requests to Dr. E. R. Chung, Department of Dairy Science, College of Agriculture, Sang Ji University, Wonju 220-702, Korea.

[2] Department of Dairy Science, College of Animal Husbandry, Kon Kuk University, Seoul 133-701, Korea.

Received June 13, 1994
Accepted December 10, 1994

determined in part by the frequencies of the alleles (Falconer, 1989). Milk protein genetic variants differ from breed to breed in their occurrence and frequency.

Thus, information is required on the gene frequencies of milk protein genetic variants in the population. The frequencies of marker genes which determine protein variants in cattle can also be used to indicate the ancestral origins and the genetic relationships between cattle breeds (Rendel, 1967; Kidd and Pircher, 1971; Kidd, 1974; Baker, 1982; Graml and Pirchner, 1984; Gonzalez et al., 1987).

The Korean native cattle (*Bos taurus chosenus*) are unique indigenous cattle breed raised in Korea. The origin of the Korean native cattle is considered to be crossbred of *Bos primigenius* and *Bos indicus*. Korea began to raise cattle about 1,800-2,000 years ago and those cattle were supposed to be brought to Korea by nomadic people from China. The first export of Korean native cattle was made to Japan during the period of the Koryo Dynasty (A.D. 918-1,392). From the gene frequency data of blood groups and serum protein polymorphism of East Asian cattle, it has been demonstrated that the Korean native cattle have greatly contributed to the gene pool of the Japanese native cattle, especially the Japanese brown cattle (Abe et al., 1968; Namikawa, 1972). The Korean native cattle now play an important role as a beef producer and as a genetic resource in Korea.

The objective of this study was to evaluate milk protein genetic polymorphisms as genetic marker in the Korean native cattle, and to compare the milk protein gene frequencies of the Korean native cattle with those of imported cattle breeds raised in Korea and Japanese brown cattle. In addition, this paper deals with the finding of a new β-casein variant.

Materials and Methods

Milk samples

Individual milk samples from a total of 280 cows were randomly collected from Alpine Experiment Station and commercial farms of Kang Won Do district. Skim milk was separated from the fat by centrifugation of whole milk at 4℃. The skim milk was stored frozen at −20℃ until electrophoresis.

Genetic marker typing

Milk proteins examined in this study were as follows; αs_1-CN, β-CN, κ-CN and β-LG. Simultaneous phenotyping of αs_1-CN, β-CN, κ-CN and β-LG genetic variants was performed by slightly modified technique of thin layer of starch urea gel electrophoresis originally described by Aschaffenburg and Thymann (1965) and Aschaffenburg and Michalak (1968). Resolution of β-CN A variant into A^1, A^2 and A^3 was achieved by starch gel electrophoresis using acid buffer system described by Arave (1967) with a slight modification.

Statistical analysis

Gene frequencies at the four milk protein loci were determined by simple gene counting method (Pirchner, 1983). The genotype distribution within codominant systems was examined for the Hardy-Weinberg equilibrium by chi-square tests (Falconer, 1989). Comparison of gene frequencies for the four milk protein loci between breeds was accomplished using two-way contingency tables (Steel and Torrie, 1980).

Results and Discussion

Starch gel electrophoretic method was used to separate the genetic variants of αs_1-CN, β-CN, κ-CN and β-LG in Korean native cattle. The electrophoretic patterns of αs_1-CN, β-CN, κ-CN and β-LG genetic variants separated on alkaline urea gel are presented in figure 1 and 2. The good separation of the β-LG bands from the leading β-CN band is shown in figure 2. Figure 3 and 4 show the electrophoretic patterns to resolve the β-CN A genetic variants on acid urea gels.

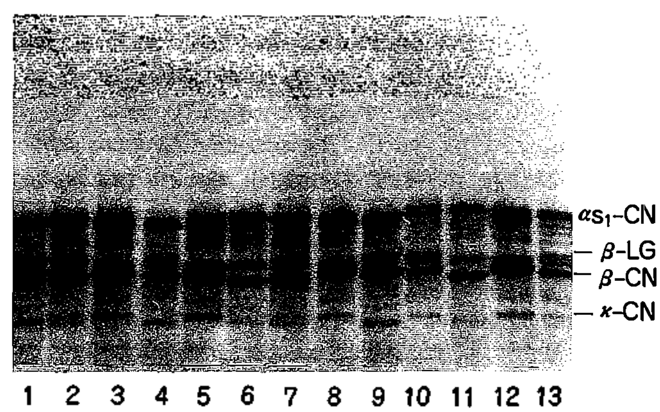

Figure 1. Starch urea gel electrophoresis at alkaline pH. Genotypes at the αs_1-CN, β-CN, κ-CN and β-LG loci are, respectively: 1) CC /AA /BB /AA, 2) BB /AB /AB /AB, 3) BC /AA /AA /BB, 4) CC /AA /BB /AA, 5) BC /AA /AA /BB, 6) BB /BB /BB /BB, 7) BB / AB /AB /AB, 8) BC /AA /AA /BB, 9) CC / AA /BB /AA, 10) BB /AA /AA /AB, 11) BB /BB /BB /BB, 12) BC /AA /AA /BB, 13) BB /AA /AA /AB.

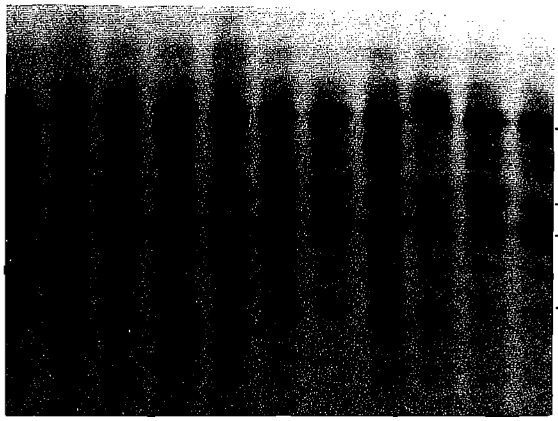

Figure 2. Starch urea gel electrophoresis showing the classification of β-LG genotypes at alkaline pH. Genotypes for β-LG locus are, respectively: 1) BB, 2) AB, 3) AA, 4) AB, 5) AB, 6) AA, 7) AA, 8) BB, 9) AB, 10) AB, 11) BB.

TABLE 1. GENE FREQUENCIES AND OBSERVED NUMBER OF THE αs_1-, β-, κ-CN AND β-LG LOCI IN KOREAN NATIVE CATTLE

Locus	Allele	Frequency	SE[1]	No.
αs_1-CN	B	.846	.015	474
	C	.154	.015	86
β-CN	A^1	.216	.017	121
	A^2	.666	.020	373
	A^4	.048	.009	27
	B	.070	.011	39
κ-CN	A	.648	.020	363
	B	.352	.020	197
β-LG	A	.148	.015	83
	B	.852	.015	477

[1]Standard error.

TABLE 2. GENOTYPE DISTRIBUTION AND HARDY-WEINBERG EQUILIBRIUM TEST FOR THE FOUR MILK PROTEIN LOCI IN KOREAN NATIVE CATTLE

Locus	Genotype	No. of animals		χ^2	df	P
		Observed	Expected			
αs_1-CN	BB	203 (72.5)[1]	200.4			
	BC	68 (24.3)	73.0	1.249	1	< .50
	CC	9 (3.2)	6.6			
β-CN	A^1A^1	9 (3.2)	13.1			
	A^2A^2	115 (41.1)	124.2			
	A^1A^2	94 (33.6)	80.6			
	A^1A^4	5 (1.8)	5.8			
	A^2A^4	19 (6.8)	17.9	8.085	5	< .25
	BA^1	4 (1.4)	8.5			
	BA^2	30 (10.7)	26.1			
	BA^4	3 (1.1)	1.9			
	BB	1 (.3)	1.4			
κ-CN	AA	116 (41.4)	117.6			
	AB	131 (46.8)	127.7	.190	1	< .75
	BB	33 (11.8)	34.7			
β-LG	AA	6 (2.1)	6.1			
	AB	71 (25.4)	70.6	.004	1	< .95
	BB	203 (72.5)	203.3			

[1]Numbers in parentheses represent percentage.

Gene frequencies for genetic variants of the four milk protein loci are summarized in table 1, and genotypic frequencies are shown in table 2. In this study, only two αs_1-CN variants, B and C, were found in milk samples of Korean native cattle. The αs_1-CN A variant occurs in very low frequency in both Holstein (Thompson et al., 1962) and Red Danish cattle (Farrell et al., 1971). The αs_1-CN D variant was discovered in Flamande cattle, and shown to occur in low frequency (Grosclaude et al., 1966). The rare αs_1-CN E variant occurred in the milk of Yak and Bali cattle (Grosclaude et al., 1976; Bell et al., 1981). The predominant gene of αs_1-CN in Korean native cattle was αs_1-CN B with a frequency of .846, whereas αs_1-CN C was in low frequency (.154). The αs_1-CN B variant is predominant in *Bos taurus,* but αs_1-CN C is in *Bos indicus* (Aschaffenburg, 1968). In the genotypic frequencies, homozygous BB accounted for 72.5% of the population followed by heterozygous BC with 24.3% and homozygous CC with 3.2%. Seven genetic variants of β-CN are known so far. These variants differ by one or more charged amino acids (Eigel et al., 1984), which makes it possible to separate the β-CN variant. The known genetic variants of β-CN are more numerous than those of the other caseins and their differentiation by gel electrophoresis were complicated. In alkaline gels, they migrate in the order $A^1 = A^2 = A^3 > B > D, E > C$. However, in acid gels (Kiddy, 1975), their order is $C > B = D > A^1 = E > A^2 > A^3$. Thus, although A variants can be differentiated from B, C, D and E by electrophoresis under alkaline conditions, acidic conditions are required for differentiation of A variants. Among the seven known genetic variants of β-CN, three genetic variants, A^1, A^2 and B, were observed. In addition, as shown in figure 3 and 4, a new variant of β-CN was found in Korean native cattle. This new β-CN band had a much slower electrophoretic mobility than the β-CN A^3 band in acid gel electrophoresis. Hence the allele controlling it was designated β-CN A^4. This band had the same colour as β-CN when stained with Amido Black 10 B in the same way as did β-CN A^1, A^2, A^3 and B. The new β-CN A^4 variant does not seem to be the same as β-CN C, D and E according to comparisons of measured distances of the migration. Aschaffenburg (1968) proposed a multiple allele theory of inheritance with the types controlled by β-CN A^1, A^2, A^3, B and C. Enough family data were not available to study the inheritance of β-CN A^4. However, according to the phenotypic exhibition of 3 heterozygote types, β-CN A^1 A^4, A^2 A^4 and BA^4, the occurrence of an additional β-CN allele is assumed. And its frequency within the population was estimated to be .048. Milk protein gene can be subject to a deletion, a duplication or an insertion. In some cases this leads to a change in the amino acid composition of the protein, resulting in a new genetic variant. There is a variation in the histidine content of β-CN A, found by sequencing the variants of β-CN. Using β-CN A^2 as the base sequence, β-CN A^1 has a histidine replacing proline at position 67, and β-CN A^3 has a glutamine replacing histidine at position 106 (Eigel et al., 1984). Thus it will be interesting to compare the amino acid composition and the primary structure of β-CN A^4 variant with those of β-CN A^1, A^2 and A^3 variants. The informations on the genetic control and physicochemical background for very slow mobility of the new variant are not yet available. Therefore, further work on the genetic and biochemical mechanisms of this new β-CN A^4 variant is in progress. On the other hand, a new β-CN A^4 variant seems to be the same as β-CN A^1 reported by Abe et al. (1975) in Japanese brown cattle, according to the comparisons of measured distances of the migration on photograph. In the β-CN locus, the A^2 allele is predominant (.666) and the B allele appears at a relatively low frequency (.070). Both Aschaffenburg (1968) and Li and Gaunt (1972) studied the β-CN A^1, A^2, A^3, B and C gene frequencies in the

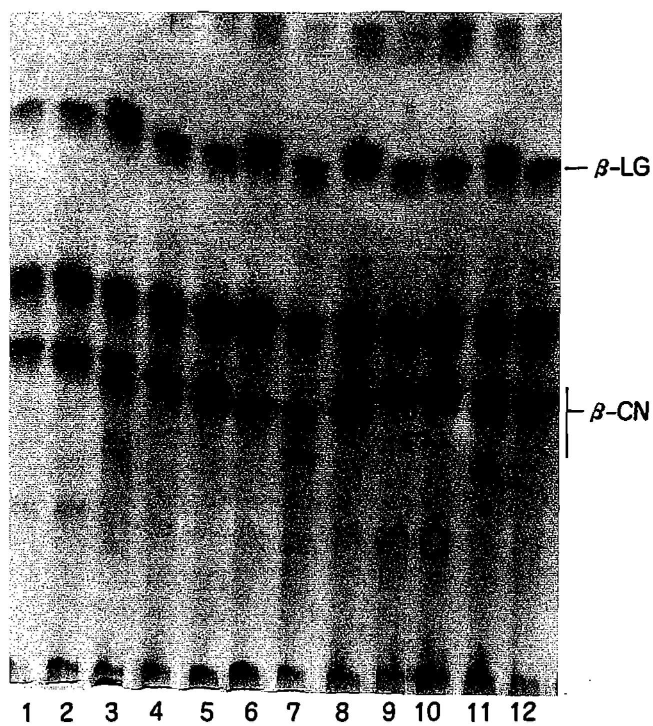

Figure 3. Starch urea gel electrophoresis at acid pH. Genotypes at β-CN locus are, respectively: 1) BB, 2) BA^1, 3) BA^2, 4) A^1A^1, 5) A^1A^2, 6) $A^1{}_2{}^2$, 7) A^2A^4, 8) A^1A^2, 9) BB, 10) BA^1, 11) BA^2, 12) A^1A^1.

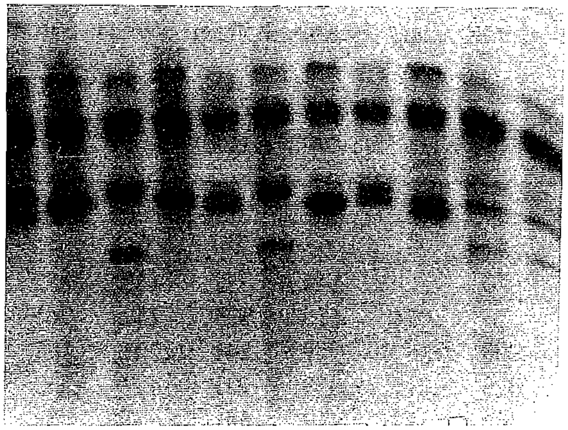

Figure 4. Starch urea gel electrophoresis at acid pH. Samples from lanes 1 and 9 are Holstein milk. Genotypes at β-CN locus are, respectively: 1) A^1A^3, 2) A^2A^2, 3) BA^4, 4) A^1A^1, 5) A^1A^2, 6) A^1A^4, 7) A^2A^2, 8) A^1A^2 9) A^2A^3, 10) A^2A^4, 11) A^2A^4.

five major Western dairy breeds. In both studies. β-CN A^1, A^2 and B accounted for over 95% of the β-CN, β-CN A^2 occurs most often, followed by variants A^1, B, C and A^3. The β-CN D variant was discovered in the milk of a few Deshi and Boran cows (Aschaffenburg, 1968) and the β-CN E in Italian Piedmont cattle (Voglino, 1972). Among the nine different genotypes of β-CN locus, the most predominant genotypes were homozygous A^2A^2 and heterozygous A^1A^2 with frequencies 41.1 and 33.6. The majority of the A^2 allele for β-CN was found in combination with the A^1 allele. For β-CN B variant, there were .3% homozygous for B and heterozygotes of combinations of A^1, A^2 and A^4 accounted for 13.2% of the β-CN. The new β-CN A^4 allele appeared only as heterozygous A^1A^4, A^2A^4, BA^4 with frequencies 1.8, 6.8 and 1.1%, respectively. In the κ-CN locus, two κ-CN alleles, A and B, were found in Korean native cattle. However, the κ-CN C and E alleles were reported by Putz et al. (1991) in Simmental cattle. The κ-CN A allele was observed with a frequency of .648, but κ-CN B allele showed to occur in low frequency (.352). The κ-CN A allele tends to be predominant in most breeds except in the Jersey, Normande and some South African zebu cattle (Aschaffenburg, 1968). For genotypic frequencies, there were 46.8% for heterozygous AB, 41.4% for homozygous AA and 11.8% for homozygous BB. Among the nine alleles for β-LG, two alleles, β-LG A and B, were observed in Korean native cattle, whereas the C, D, E, F, G, H and W alleles of β-LG were not identified. The predominant β-LG allele was β-LG B with .852 frequency, while allele frequency of β-LG A was very low (.148). The predominant genotype of β-LG locus was homozygous BB with 72.5% frequency. There were 25.4 % heterozygous AB but only 2.1% homozygous AA for β-LG. As a consequence, in Korean native cattle population the most prevalent alleles were B for αs_1-CN, A for β-CN (A^2 for the subset), A for κ-CN and B for β-LG. The observed and expected genotype frequencies showed no significant differences (table 2). According to the Hardy-Weinberg law, the Korean native cattle populations were in genetic equilibrium.

Gene frequencies for genetic variants of the four milk protein loci were compared with those reported in the literature for imported cattle breeds raised in Korea and Japanese brown cattle (table 3 and 4). The Korean native cattle population showed significant differences in the different milk protein gene frequencies compared with those of Holstein breed. Significant differences in gene frequencies between the Korean native cattle and the three imported beef cattle breeds were also observed at milk protein loci, with the exception of αs_1-CN locus in Charolais and β-LG locus in Augus. Therefore, the genetic constitution of the Korean native cattle was considered to deviate remarkably from those of imported cattle breeds raised in Korea. However, gene frequencies in Korean native cattle were very similar to those reported in the literature for the Japanese brown cattle. There were no significant differences in gene frequencies for milk protein loci between two breeds, except for β-CN locus. The β-CN B allele was found in Korean native cattle, whereas the β-CN C allele in Japanese brown cattle. The β-CN A^1 allele had the higher frequency in Korean native cattle, while the β-CN A^2 allele in Japanese brown cattle. Interestingly, the new β-CN A^4 gene which is supposed to be one of the characteristics of Korean native cattle, exhibits an extremely low frequency (.011) in the Japanese brown cattle. This supports the idea that there has been a genetic connection between the two breeds. The occurrence of this new β-CN A^4 allele in the Japanese brown cattle can be explained that Korean native cattle were used in the development of the Japanese brown cattle. From the data using the frequencies of blood groups and serum protein polymorphisms, Abe et al. (1968) reported that the Korean native cattle have contributed to the gene pool of the Japanese brown cattle. Also, Ito et al. (1988) reported that the Japanese brown cattle had a closer genetic relationship with the Korean native cattle than with Simmental using the frequencies of 17 blood groups and blood protein loci. Results from this study provide a good evidence for supporting the previous studies. Consequently, this study and others indicate that the genetic constitution of the Korean native cattle were very similar to those of the Japanese brown cattle, and Japanese brown cattle originated from Korean native cattle.

TABLE 3. GENE FREQUENCIES FOR THE FOUR MILK PROTEIN LOCI IN SIX DIFFERENT CATTLE BREEDS

Breed	No. of animals	αs_1-CN		β-CN						κ-CN		β-LG	
		B	C	A^1	A^2	A^3	A^4	B	C	A	B	A	B
KNC[1]	280	.846	.154	.216	.666	–	.048	.070	–	.648	.352	.148	.852
Jap. brown[2]	88	.813	.187	.136	.818	–	.011	–	.034	.625	.375	.239	.761
Angus[3]	86	.942	.058	.228	.766	–	–	.006	–	.759	.241	.203	.797
Hereford[3]	92	.967	.033	.511	.450	–	–	.028	.011	.879	.121	.457	.543
Charolais[3]	32	.891	.109	.194	.371	–	–	.306	.129	.419	.581	.641	.359
Holstein[4]	138	.949	.051	.471	.460	.040	–	.029	–	.728	.272	.446	.554

[1] Korean native cattle.
[2] Japanese brown cattle; Data from Abe et al. (1975).
[3] Data from Chung et al. (1993).
[4] Data from Han et al. (1984[a,b]).

TABLE 4. COMPARISONS OF GENE FREQUENCIES BETWEEN KOREAN NATIVE CATTLE AND FOREIGN CATTLE BREEDS FOR THE FOUR MILK PROTEIN LOCI

Breed	df	χ^2	P	df	χ^2	P
		αs_1-casein			β-casein	
KNC[1] vs. Jap. brown[2]	1	1.14	<.50	4	44.68	<.001
KNC vs. Angus	1	10.52	<.001	3	20.03	<.001
KNC vs. Hereford	1	18.70	<.001	4	71.29	<.001
KNC vs. Charolais	1	.89	<.50	4	116.79	<.001
KNC vs. Holstein	1	18.57	<.001	4	94.51	<.001
		κ-casein			β-lactoglobulin	
KNC vs. Jap. brown	1	.31	<.75	1	1.55	<.25
KNC vs. Angus	1	7.71	<.01	1	.30	<.75
KNC vs. Hereford	1	35.96	<.001	1	21.22	<.01
KNC vs. Charolais	1	12.55	<.001	1	87.46	<.001
KNC vs. Holstein	1	5.40	<.025	1	88.08	<.001

[1] Korean native cattle.
[2] Japanese brown cattle.

Literature Cited

Aaltonen, M. L. and V. Antila. 1987. Milk renneting properties and the genetic variants of proteins. Milchwissenschaft 42(8):490-492.

Abe, T., M. Komatsu, T. Oishi and A. Kageyama. 1975. Genetic polymorphism of milk proteins in Japanese cattle and European cattle breeds in Japan. Jap. J. Zootech. Sci. 46(1):591-599.

Abe, T., T. Oishi, S. Suzuki, T. Amano, K. Kondo, K. Nozawa, T. Namikawa, K. Kumazaki, O. Koga, S. Hayashida and J. Otsuka. 1968. Studies on the native farm animals in Asia. I. On the blood groups and serum protein polymorphism of East Asian cattle. Jap. J. Zootech. Sci. 39(12):517-522.

Aleandri, R., L. G. Buttazzoni, J. C. Schneider, A. Caroli and R. Davoli. 1990. The effects of milk protein polymorphisms on milk components and cheese producing ability. J. Dairy Sci. 73:241-255.

Arave, C. W. 1967. Procedures for simultaneous phenotyping of β-casein and β-lactoglobulin variants in cow's milk. J. Dairy Sci. 50:1320-1322.

Aschaffenburg, R. 1968. Review of the progress of dairy science. Section G. Genetic variants of milk proteins: their breed distribution. J. Dairy Res. 35:447-460.

Aschaffenburg, R. and J. Drewry. 1955. Occurrence of different β-lactoglobulins in cow's milk. Nature (London) 176:218-219.

Aschaffenburg, R. and W. Michalak. 1968. Simultaneous phenotyping procedures for milk proteins. J. Dairy Sci. 51:1849-1952.

Aschaffenburg, R. and M. Thymann. 1965. Simultaneous phenotyping procedure for the principal proteins of cow's milk. J. Dairy Sci. 48:1524-1526.

Baker, C. M. A. 1982. The use of genetic relationships among cattle breeds in the formulation of rational breeding policies: A re-examination of the example of the South Devon and the Gelbvieh. Anim. Blood Grps biochem. Genet. 13:199-212.

Bech, A. M. and K. R. Kristiansen. 1990. Milk protein polymorphism in Danish dairy cattle and the influence of genetic variants on milk yield. J. Dairy Res. 57:53-62.

Bell, K., H. A. McKenzie and D. C. Shaw. 1981. Bovine β-lactoglobulin E. F and G of Bali (banteng) cattle, Bos (Bibos) javanicus. Aust. J. Biol. Sci. 34:133-147.

Bovenhuis, H., J. A. M. V. Arendonk and S. Korver. 1992. Associations between milk protein polymorphisms and milk production traits. J. Dairy Sci. 75:2549-2559.

Chung, E. R., C. H. Yu, H. Y. Chung, J. E. Kim, K. J. Chun, S. K. Han and Y. C. Shin. 1993. Studies on polymorphisms of milk proteins as genetic markers in beef cattle. Korean J. Anim. Sci. 35(3):181-190.

Eenennaam, A. V. and J. F. Medrano. 1991. Milk protein polymorphisms in California dairy cattle. J. Dairy Sci. 74:1730-1742.

Eigel, W. N., J. E. Butler, C. A. Ernstrom, H. M. Farrell JR, V. R. Harwalker, R. Jenness and R. McL. Whitney. 1984. Nomenclature of proteins of cow's milk:Fifth revision. J. Dairy Sci. 67:1599-1631.

Falconer, D. S. 1989. Introduction to quantitative genetics (3rd. ed.). Longman Scientic & Technical. pp. 4-50.

Farrell, JR, H. M., M. P. Thompson and B. Larsen. 1971. Verification of the occurrence of the αs_1-casein A allele in Red Danish cattle. J. Dairy Sci. 54(3):423-425.

Gonyon, D. S., R. E. Mather, H. C. Hines, G. F. W. Haenlein, C. W. Arave and S. N. Gaunt. 1987. Associations of bovine blood and milk polymorphisms with lactation traits:Holsteins. J. Dairy Sci. 70:2585-2598.

Gonzalez, P., M. J. Tunon and M. Vallejo. 1987. Genetic relationships between seven Spanish native breeds of cattle. Anim. Genet. 1987(18):249-256.

Graml, R., J. Buchberger, H. Klostermeyer and F. Pirchner. 1986. Pleiotrope wirkungen von β-lactoglobulin und casein genotypen auf milchfett und milchproteinmengen des bayerischen Flekviehs und Braunviehs. J. Anim. Breedg. Genet. 103:33-45.

Graml, R. and F. Pirchner. 1984. Relation of genetic distance between cattle breeds and heterosis of resulting crosses. Anim. Blood Grps. biochem. Genet. 15:173-180.

Grosclaude, F. 1988. Genetic polymorphism of the principal milk proteins in cattle. Relations with quantity, composition and cheesemaking suitability of milk. INRA Productions Animales 1:5-17.

Grosclaude, F., M. F. Mahe, J. C. Mercier, J. Bonnemaire and J. H. Teissier. 1976. Polymorphisme de lactoproteines de bovines nepalaise. Ann. Genet. Sel. Anim. 8:461-479.

Grosclaude, F., J. Pujolle, B. Ribadeau-Dumas and J. Garnier. 1966. Analyse genetique du groupe de loci de structure synthetisant les caseines bovines. Xth European Conference on Animal Blood Groups and Biochemical Polymorphism. Paris. 415-420.

Haenlein, G. F. W., D. S. Gonyon, R. E. Mather and H. C. Hines. 1987. Associations of bovine blood and milk polymorphisms with lactation traits: Guernseys. J. Dairy Sci. 70:2599-2609.

Han, S. K., K. M. Lee, E. R. Chung and K. J. Jang. 1984[a]. Studies on the genetic polymorphism in milk proteins. Ⅰ. Genetic variants of αs_1-casein and κ-casein. Korean J. Anim. Sci. 26(3):212-216.

Han, S. K., K. M. Lee, E. R. Chung and K. J. Jang. 1984[b]. Studies on the genetic polymorphism in milk proteins. Ⅱ. Genetic variants of β-casein and β-lactoglobulin. Korean J. Anim. Sci. 26(3):217-224.

Hargrove, G. L., C. A. Kiddy, C. W. Young, A. G. Hunter, G. W. Trimberger and R. E. Mather. 1980. Genetic polymorphisms of blood and milk and reproduction in Holstein cattle. J. Dairy Sci. 63:1154-1166.

Ito, S. I., M. Kanemaki, M. Morita, M. Yamada, Y. Tanabe, T. Nagamura, T. Namikawa and T. Tomita. 1988. Blood protein and blood group gene constitutions of Japanese brown cattle in Kumamoto and their genetic relationships with Korean and Simmental cattle. Jpn. J. Zootech. Sci. 59(5):433-445.

Jairam, B. T. and P. G. Nair. 1983. Genetic polymorphisms of milk proteins and economic characters in dairy animals. Ind. J. Anim. Sci. 53:1.

Kidd, K. K. 1974. Biochemical polymorphisms, breed relationships and germ plasm resources in domestic cattle. Proceedings of the 1st World Congress on Genetic Applied to Livestock Production, Madrid 1:321-328.

Kidd, K. K. and F. Pirchner. 1971. Genetic relationships of Austrian cattle breeds. Anim. Blood Grps. biochem. Genet. 2:145-158.

Kiddy, C. A. 1975. Gel electrophoresis in vertical polyacrylamide beds. Procedure 1. In: H. Swaisgood (Ed.). Methods of gel electrophoresis of milk proteins. Dept. of Science N. C. State Univ., Raleigh, USA. pp. 14-19.

Li, F. H. G. and S. N. Gaunt. 1972. A study of genetic polymorphisms of milk β-lactoglobulin. αs_1-casein, β-casein and κ-casein in five dairy breeds. Biochem. Genet. 6:9-20

Lin, C. Y., A. J. McAllister, K. F. Ng-Kwai-Hang and J. F. Hayes. 1986. Effects of milk protein loci on first lactation production in dairy cattle. J. Dairy Sci. 69:704-712.

Lin, C. Y., A. J. McAllister, K. F. Ng-Kwai-Hang, J. F. Hayes, T. R. Batra, A. J. Lee, G. L. Roy, J. A. Vesely, J. M. Wauthy and K. A. Winter. 1987. Association of milk protein types with growth and reproductive performance of dairy heifers. J. Dairy Sci. 70:29-39.

Lin, C. Y., A. J. McAllister, K. F. Ng-Kwai-Hang, J. F. Hayes, T. R. Batra, A. J. Lee, G. L. Roy, J. A. Vesely, J. M. Wauthy and K. A. Winter. 1989.

Relationships of milk protein types to lifetime performance. J. Dairy Sci. 72:3085-3090.

Marziali, A. S. and K. F. Na-Kwai-Hang. 1986. Relationships between milk protein polymorphisms and cheese yielding capacity. J. Dairy Sci. 69:1193-1201.

McLean, D. M. 1987. Influence of milk protein genetic variants on milk composition, yield and cheesemaking properties. Anim. Genet. 18 (Suppl. 1):100-102.

McLean, D. M., E. R. B. Graham, R. W. Ponzoni and H. A. McKenzie. 1984. Effects of milk protein genetic variants on milk yield and composition. J. Dairy Res. 51:531-546.

McLean, D. W. and J. Schaar. 1989. Effects of β-lactoglobulin and κ-casein genetic variants and concentration on syneresis of gels from renneted heated milk. J. Dairy Res. 56:297-301.

Namikawa, T. 1972. Genetic similarities among seven cattle populations of eastern Asia and Holstein breed. SABRAO Newsletter 4(1):17-25.

Ng-Kwai-Hang, K. F., J. F. Hayes, J. E. Moxley and H. G. Monardes. 1984. Association of genetic variants of casein and milk serum proteins with milk, fat and protein production by dairy cattle. J. Dairy Sci. 67:835-840.

Ng-Kwai-Hang, K. F., J. F. Hayes, J. E. Moxley and H. G. Monardes. 1986. Relationships between milk protein polymorphisms and major milk constituents in Holstein-Friesian cows. J. Dairy Sci. 69:22-26.

Pagnacco, G. and A. Caroli. 1987. Effect of casein and β-lactoglobulin genotypes on renneting properties of milks. J. Dairy Res. 54:479-486.

Pirchner, F. 1983. Population Genetics in Animal Breeding (2nd ed.). Plenum Press, New York and London, pp. 6-12.

Putz, M., G. Averdunk, J. Aumann and J. Buchberger. 1991. Genotypen fur milch-protein beim bayerischen Fleckvieh. Der Tierzuchter 43:479.

Rendel. 1967. Studies of blood groups and protein variants as a means of revealing similarities and differences between animal populations. Animal Breeding Abstr. 35(3):371-383.

Ronda, R. and O. Perez-Beato. 1983. Relationship between polymorphic proteins and production and reproductive characters in Red-and-White Holstein cows. 2. Milk proteins. Anim. Breed. Abstr. 51:808.

Schaar, J., B. Hansson and H. E. Petterson. 1985. Effects of genetic variants of κ-casein and β-lactoglobulin on cheesemaking. J. Dairy Res. 52:429-437.

Singh, H., P. N. Bhat and R. Singh. 1981. Association of protein polymorphic genotypes with certain performance traits in crossbred cattle. Ind. J. Anim. Sci. 51:5.

Steel, R. G. and J. H. Torrie. 1980. Principles and procedures of statistics. a biomedical approach (2nd ed.). McGraw-Hill, Inc. pp. 469-520.

Thompson, M. P., C. A. Kiddy, L. Pepper and C. A. Zittle. 1962. Variations in the αs-casein fraction of individual cow's milk. Nature 195:1001.

Voglino, G. F. 1972. A new β-casein variant in Piedmont cattle. Anim. Blood Grps biochem. Genet. 3:61-62.