

The Relationship between Milk Protein Phenotypes and Lactation Traits in Brown Swiss and Canadienne

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ABSTRACT: A total of 1033 Brown Swiss and 610 Canadienne cows were phenotyped for the genetic variants α_{s1} -casein, β -casein, κ -casein, β -lactoglobulin and α -lactalbumin. In Brown Swiss, frequency distributions were: 97.3% B and 2.7% C variant of α_{s1} -casein; 31.6% A¹, 51.8% A², 0.5% A³ and 16.1% B variant of β -casein; 70.4% A, 29.3% B, and 0.3% C variant of κ -casein; 41.7% A and 58.3% B variant of β -lactoglobulin; and 100% B variant of α -lactalbumin. Corresponding frequencies in Canadienne for those five milk proteins were: 98.6 and 1.4%; 58.5, 33.5, 0.08 and 7.9%; 78.8, 21.1 and 0.1%, 42.4 and 57.6%; and 100%. Analysis of variance by least squares showed possible association between milk protein phenotypes and some

lactational production traits. There were no significant association of phenotypes of α_{s1} -casein, β -casein and β -lactoglobulin with milk yield, fat yield, protein yield, fat percentage and protein percentage in both breeds during the three lactations. In the Brown Swiss, κ -casein phenotype was associated with 305-day fat yield and protein yield during the first lactation. κ -Casein AB was associated with higher milk, fat and protein yields during the second lactation. During the third lactation, β -lactoglobulin AA in Canadienne cows was associated with higher protein content in the milk (3.70%) when compared to phenotypes AB (3.54%) and BB (3.64%).

(**Key Words:** Brown Swiss, Canadienne, Genetic Variants, milk, Fat, Protein)

INTRODUCTION

The major milk proteins are polymorphic and it has been demonstrated that frequency distributions of the genetic variants are breed specific (Aschaffenburg, 1968; Grosclaude, 1988; Buchberger, 1995). Ng-Kwai-Hang and Grosclaude (1992) reported that numerous studies have indicated close associations between certain milk protein variants and milk production traits. The results obtained in many of those studies are conflicting because different breeds of cattle were under consideration. Most of the reports on cow populations that are large enough to permit meaningful statistical analysis of the data are confined to the Holstein breed (Ng-Kwai-Hang et al., 1984, 1986, 1990; Gonyon et al., 1987; Aleandri et al., 1990; Bovenhuis et al., 1992). In studies of associations between milk protein genetic variants and production traits it is important to assess whether the relationships are breed specific. Our earlier investigation (Kim et al., 1996), reported on the relationship between milk protein phenotypes and production traits in Ayrshires and Jerseys. The present study on genetic polymorphism and production traits in the Quebec Brown Swiss and Canadienne breeds are similar to that reported earlier

(Kim et al., 1996) on Ayrshires and Jerseys. Hence, the objective of this project was to investigate possible associations between genetic variants of α_{s1} -casein, β -casein, κ -casein and β -lactoglobulin and lactational production of milk, fat and protein for three lactations in Brown Swiss and Canadienne.

MATERIALS AND METHODS

Data collection for lactation traits

Lactation production records including milk yield, fat yield, protein yield, fat (%) and protein (%) during the first, second and third lactation were obtained from the Quebec Dairy Herd Analysis Service. In all, there were 30 Brown Swiss and 14 Canadienne herds registered in the milk recording program. On an approximately monthly basis, milk productions from individual cows were recorded for the morning and afternoon milkings. A sample of the combined milkings was sent to the laboratory for analysis of fat and protein with an infrared Multispec II (Multispec Inc. Wheldrake, York, England) milk analyser which was previously calibrated with milk standards according to the manufacturer's specifications. The monthly test-day analyses for fat and protein

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contents were used to calculate the values on a lactational basis according to established standard procedures employed in genetic evaluations in Canada. Briefly, the average of adjacent test-day yield is multiplied by the number of days between the adjacent test days and then summed over all test-day intervals during the given lactation. From the production files, data pertaining to test-day, lactation number, calving date, fat and protein contents, 305 d lactation milk, fat and protein yields were retrieved for the individual cows.

Phenotyping of milk proteins

Polyacrylamide gel electrophoresis under alkaline and reducing conditions were used to determine the phenotypes of α -_{s1}-casein, β -casein and κ -casein (Ng-Kwai-Hang et al., 1984). To differentiate between variants A¹, A² and A³ of β -casein it was necessary to run the electrophoretic separation under acidic conditions (Ng-Kwai-Hang et al., 1984). The electrophoretic method of Ng-Kwai-Hang and Kroeker (1984) was used to characterise the genetic variants of β -lactoglobulin and α -lactalbumin. To avoid errors due to misidentification or mix-up of samples the phenotyping procedures were repeated on milk samples collected on three different occasions. The results obtained for individual cows were compared and retained only if they were consistent for the three occasions. A file was created to match each cow with its phenotype for α -_{s1}-casein, β -casein, κ -casein, β -lactoglobulin and α -lactalbumin.

Statistical analyses

Because of differences in frequency distributions of milk protein variants and differences in production traits for Brown Swiss and Canadienne, analyses were carried out separately for the two breeds. The associations of milk protein phenotypes with milk, fat and protein yields and percentages of fat and protein were analysed separately according to lactation number. The least squares model used was according to SAS (1990) and included herd, age of cow at calving, year and month of calving, phenotype of α -_{s1}-casein, β -casein, κ -casein and β -lactoglobulin.

After matching complete sets of production data in Brown Swiss, there remained 145, 119 and 86 cows for the first, second and third lactation, respectively. There were 15, 18, and 18 subclasses for age at calving in the three respective lactations. Each of the subclasses for age of calving were classified on a monthly interval and represented cows which calved within the age of 24 to 38 mo for the first lactation, 35 to 52 mo for the second lactation and 49 to 66 mo for the third lactation. For all three lactations, there were 6 subclasses of six months

each, for season of calving which occurred from September 1989 to August 1992. In the Brown Swiss population, there were three phenotypes for α -_{s1}-casein: -BB, BC, CC. Because only 0.10% of the cows were of phenotype CC, records with this classification for α -_{s1}-casein type were removed for statistical analysis. Thus only two types of α -_{s1}-casein (BB and BC) were considered. Phenotypes for β -casein were divided into six subclasses corresponding to A¹A¹, A¹A², A²A², A¹B, A²B and BB. There were three subclasses (AA, AB, BB) each for κ -casein and β -lactoglobulin phenotypes. Because 100% of the cows were homozygous B for α -lactalbumin, there were no subclassifications of this milk protein type.

Complete data sets were analysed for 140 cows in first lactation, 101 cows in second lactation and 63 cows in third lactation for the Canadienne breed. There were 15, 18 and 15 monthly subclasses for age at calving in the three respective lactations. The monthly subclasses for age at calving represented cows which calved at the age of 23 to 37 mo, 34 to 52 mo and 48 to 62 mo during the three respective lactations. There were five subclasses of six months each for season of calving during all three lactations. The semi-annual calving season occurred from September 1989 to August 1992. Among the Canadienne breed, 100% of the cows were of BB phenotype for α -lactalbumin and hence there was no subclassification for this protein. There were two subclasses (BB, BC) for α -_{s1}-casein. There were 5 subclasses for β -casein (A¹A¹, A¹A², A²A², A¹B, A²B) and three each for κ -casein and β -lactoglobulin (AA, AB, BB).

RESULTS AND DISCUSSION

Frequency distribution of milk protein phenotypes

Table 1 shows the frequency distribution of the phenotypes of the five milk proteins in Brown Swiss and Canadienne. All of the 1033 Brown Swiss and 610 Canadienne were of the BB phenotype for α -lactalbumin. Frequencies of the various detected phenotypes for the other four protein systems were different for the two breeds. Two variants, B and C of α -_{s1}-casein were detected in Brown Swiss and Canadienne. The frequency of variant C was low with only one individual for phenotype CC occurring in both breeds. Variant C was more frequent in Brown Swiss than in Canadienne. Phenotype BC and BB accounted for 5.13% and 94.78% in Brown Swiss compared to 2.45% and 97.38% respectively in Canadienne. A frequency of 1 to 10% for α -_{s1}-casein C and 90 to 99% for α -_{s1}-casein B in Brown Swiss has been reported in the literature (Aschaffenburg, 1968; Buchberger, 1995). The only report on the

Table 1. Phenotype frequencies of milk proteins in Brown Swiss and Canadienne

Milk protein	Phenotype	Brown Swiss		Canadienne	
		N ^o	%	N ^o	%
α_{s1} -Casein	BB	980	94.78	595	97.38
	BC	53	5.13	15	2.45
	CC	1	0.10	1	0.16
β -Casein	A ¹ A ¹	120	11.68	209	34.38
	A ¹ A ²	326	31.74	241	39.64
	A ¹ A ³	2	0.19	0	0
	A ² A ²	268	26.10	62	10.20
	A ² A ³	5	0.49	1	0.16
	A ¹ B	82	7.98	52	8.55
	A ² B	197	19.18	42	6.91
	A ³ B	3	0.29	0	0
	BB	24	2.34	1	0.16
κ -Casein	AA	489	48.04	382	62.73
	AB	449	44.11	195	32.02
	AC	6	0.58	1	0.16
	BB	74	7.27	31	5.10
β -Lactoglobulin	AA	141	13.65	95	15.57
	AB	579	56.05	327	53.61
	BB	313	30.30	188	30.82
α -Lactalbumin	BB	1,033	100	610	100

Canadienne breed (Hoogendoorn et al., 1969) indicated a frequency of 0.959 for B and 0.041 for C variant of α_{s1} -casein. Variant A¹ of β -casein with a frequency of 0.58 was more frequent in Canadienne whereas variants A², A³ and B with frequencies of 0.52, 0.005 and 0.16 were more frequent in Brown Swiss. There were no A¹A³ and A³B phenotype for β -casein in the Canadienne breed where only 0.16% of the cows were β -casein BB compared to 0.19% A¹A³, 0.29% A³B and 2.34% BB for β -casein in Brown Swiss. Frequencies of the different variants in Brown Swiss and Canadienne are within the range of values reported in literature. In a review, Buchberger (1995) reported frequencies of 0.03 to 0.24 for A¹, 0.46 to 0.70 for A², 0.15 to 0.29 for B and 0.01 to 0.06 for C variant of β -casein in Brown Swiss. An earlier study by Hoogendoorn et al. (1969) showed gene frequencies of 0.614 for A¹, 0.282 for A², 0 for A³, 0.104 for B and 0 for C variant of β -casein in Canadienne. Higher frequencies of κ -casein A than of κ -casein B were observed in Brown Swiss and Canadienne where there were 48.04 and 62.73% AA compared to 7.27 and 5.10% BB in the two respective breeds. A low frequency of κ -casein C which occurred as heterozygous AC (0.58 and 0.16%) were observed in both breeds. At the β -lactoglobulin locus, the distribution of A and B variants were similar in Brown Swiss and Canadienne. Frequencies for the three phenotypes of β -lactoglobulin

were: 13.65% AA, 56.05% AB and 30.30% BB in Brown Swiss and 15.55% AA, 53.61% AB and 30.82 BB in Canadienne.

Production data for Brown Swiss and Canadienne

Table 2 shows the 305-day complete lactation data for Brown Swiss and Canadienne which were retained for analysis after discarding records which did not have complete information with respect to phenotypes for α_{s1} -casein, β -casein, κ -casein and β -lactoglobulin, age at calving and season of calving. The lactational yields and percentages of fat and protein were within the ranges of the breeds registered in the Quebec milk recording programme (PATLQ, 1995). Brown Swiss outproduced the Canadienne, by 959, 1,535 and 1,327 kg of milk during the first, second and third lactation, respectively. In Brown Swiss, fat content increased from 3.99 to 4.15% and protein content increased from 3.51 to 3.78% from the first to the third lactation. During the same time interval, a decline in fat from 4.32 to 4.20% and in protein from 3.59 to 3.54% was observed in Canadienne. Fat and protein yields in Brown Swiss increased with increasing lactation because milk production and component compositions increased with age. Higher milk production with increased lactation number in Canadienne, overcompensated for lower component percentages as shown by higher lactational yields of fat and protein.

Table 2 Summary of 305-D lactation production of Brown Swiss and Canadienne

Breed and Lactation N°	N	Yield (kg)						Concentration (%)			
		Milk		Fat		Protein		Fat		Protein	
		Mean	SE	Mean	SE	Mean	SE	Mean	SE	Mean	SE
Brown Swiss											
1	145	5,127.2	992.0	204.6	44.8	179.5	35.8	3.99	0.37	3.51	0.22
2	119	5,999.0	1,230.5	244.7	54.7	218.8	45.3	4.07	0.32	3.55	0.21
3	86	6,432.9	1,377.9	266.5	62.8	229.7	50.5	4.15	0.41	3.78	0.23
Canadienne											
1	140	4,167.9	725.9	179.7	32.6	149.6	26.3	4.32	0.34	3.59	0.18
2	101	4,464.5	974.7	190.0	42.3	159.7	34.6	4.26	0.31	3.58	0.19
3	63	5,105.7	1,182.1	213.5	50.0	180.6	42.3	4.20	0.37	3.54	0.20

Milk protein phenotypes and production traits in Brown Swiss

Although there were three phenotypes (BB, BC, CC) for α_{s1} -casein it was only possible to determine the effects of two phenotypes, BB and BC on production traits because of the very low frequency of α_{s1} -casein CC. Similarly, only six of the nine phenotypes of β -casein were retained for analysis with production traits. Table 3 shows that α_{s1} -casein, β -casein and β -lactoglobulin phenotypes in Brown Swiss did not affect

milk, fat and protein yields and percentages of fat and protein in milk for all the three lactations. Studies involving Holsteins (Gonyon et al., 1987; Aleandri et al., 1990), Ayrshires (Hoogendoorn et al., 1969; Lin et al., 1986), Guernseys (Haenlein et al., 1987), Jerseys (Kim et al., 1996) have reported no relationships between milk yield and β -casein phenotypes. Other studies (Ng-Kwai-Hang et al., 1984, 1987, 1990; Bovenhuis et al., 1992) reported that milk production in Holsteins were associated with β -casein variants. Reports involving other breeds of

Table 3. The f ratios from least squares analyses of variance for 305-D lactation yield and milk composition for three lactations in Brown Swiss

Source	df	Milk Yield	Fat Yield	Protein Yield	Fat (%)	Protein (%)
Lactation 1						
Herd	15	4.39**	6.08**	6.24**	3.99**	5.51**
Age at calving	14	1.11	1.37	1.16	1.67	1.03
α_{s1} -Casein	1	1.97	2.12	2.42	0.19	0.15
β -Casein	5	1.04	0.96	0.73	1.43	2.13
κ -Casein	2	2.37	3.69*	3.18*	0.87	0.98
β -Lactoglobulin	2	0.81	0.14	0.47	1.44	0.73
Error	102					
Lactation 2						
Herd	15	5.02**	6.22**	6.64**	3.04**	3.13**
Age at calving	17	1.93*	1.91*	1.96*	1.57*	0.55
α_{s1} -Casein	1	0.94	0.38	0.43	0.33	1.09
β -Casein	5	0.72	0.91	0.99	1.10	0.70
κ -Casein	2	0.70	0.79	0.83	0.03	1.75
β -Lactoglobulin	2	1.24	0.94	0.84	0.16	0.26
Error	75					
Lactation 3						
Herd	14	4.14**	6.45**	5.99**	1.91	2.33**
Age at calving	17	1.67	1.64	1.76	1.42	1.21
α_{s1} -Casein	1	0.21	0.03	0.37	0.52	0.01
β -Casein	4	0.66	1.14	1.05	0.90	0.90
κ -Casein	2	2.80	1.94	2.20	2.48	1.83
β -Lactoglobulin	2	0.33	0.66	0.49	0.04	0.53
Error	44					

* $p < 0.05$, ** $p < 0.01$.

cattle (Ng-Kwai-Hang et al., 1986, 1990; Bovenhuis et al., 1992; Gonyon et al., 1987; Kim et al., 1996) have indicated significant effects of genetic variants of α_{s1} -casein, β -casein, κ -casein and β -lactoglobulin on milk fat and protein. In the present study, κ -casein phenotypes significantly ($p < 0.05$) affected fat and protein yields during the first lactation but not during subsequent lactations. No significant associations of κ -casein types with milk yield, fat percentage and protein percentages were found for the three lactations. Figure 1 shows the variation of fat yield according to κ -casein

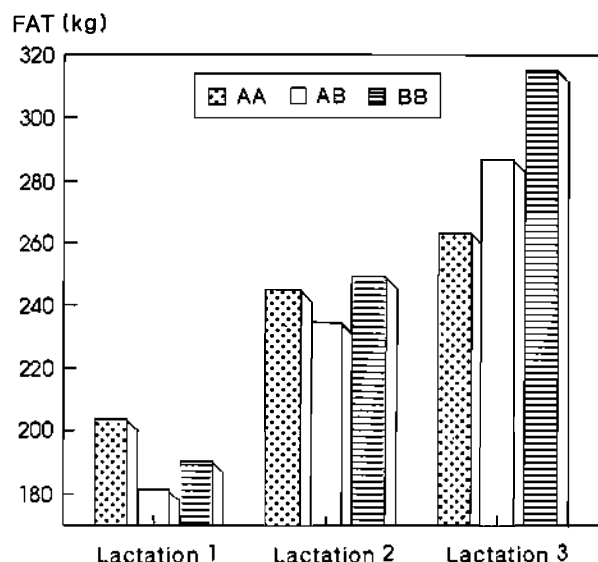


Figure 1. Association between κ -casein phenotypes and fat yield for three lactations in Brown Swiss.

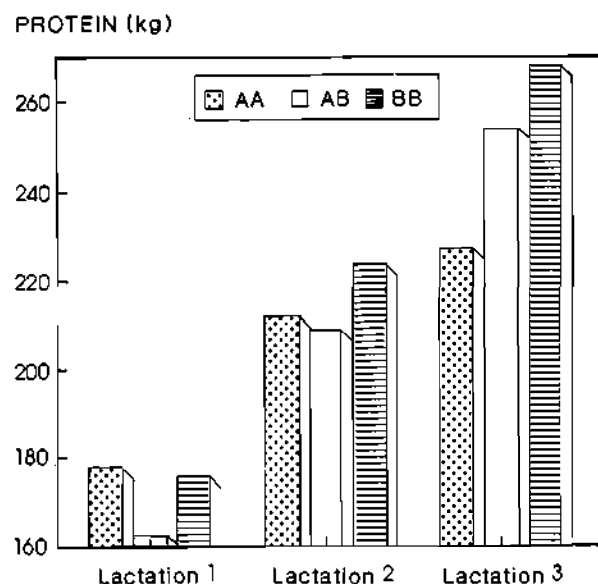


Figure 2. Association between κ -casein phenotypes and protein yield for three lactations in Brown Swiss.

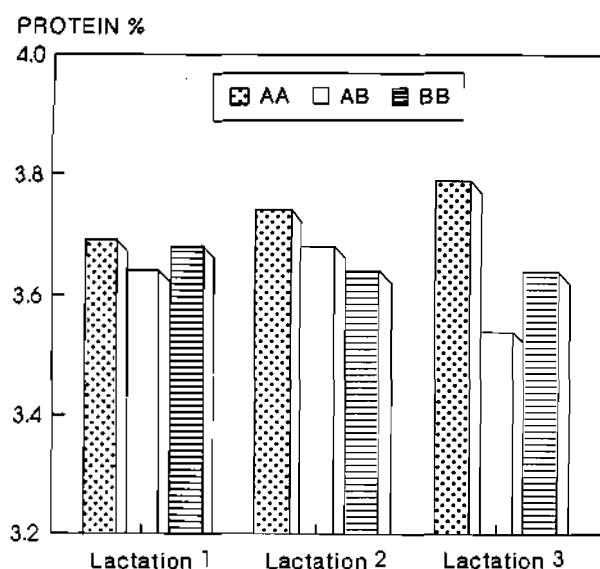
phenotype and lactation number in Brown Swiss. For the first lactation, cows with κ -casein AA produced more fat (203.5 kg) than cows with AB (181 kg) and BB (190.5 kg) phenotypes. Figure 2 shows that κ -casein AA cows produced 178.1 kg of protein compared to 162.7 kg for κ -casein AB and 175.9 kg for κ -casein BB. The patterns of fat and protein yields according to κ -casein types and lactation numbers were similar. Phenotype AA of κ -casein was associated with highest component yields during the first lactation. In contrast, κ -casein BB was associated with the highest fat and protein yields during the second and third lactations. The majority of reports for other breeds indicate that κ -casein BB and β -lactoglobulin BB are associated with higher fat content of milk whereas κ -casein BB and β -lactoglobulin AA are associated with higher protein content of milk.

Milk protein phenotypes and production traits in Canadienne

Table 4 shows no association between phenotypes of the four milk proteins and milk yield and composition for the first lactation in Canadienne. These results are consistent with an earlier study (Hoogendoorn et al., 1969) involving Canadienne and with other reports (McLean et al., 1984; Beck and Kristiansen, 1990; Kim et al., 1996; Gonyon et al., 1987; Aleandri et al., 1990; Haenlein et al., 1987) for other breeds of cattle. In the second lactation there were no effects of α_{s1} -casein, β -casein and β -lactoglobulin phenotypes on yield and composition parameters. In contrast, κ -casein types significantly ($p < 0.01$) influence milk, fat and protein yields but not fat and protein percentages. A comparison of least squares means of milk, fat and protein yields during the second lactation for the three phenotypes of κ -casein revealed that κ -casein AB was associated with higher yields than either the AA or BB phenotype. Second lactational yields were: 4,658.8 kg milk, 196.6 kg fat and 170 kg protein for κ -casein AB cows compared to 4,033.3 kg milk, 172 kg fat and 149 kg protein for κ -casein AA cows. Higher production of protein and fat for κ -casein AB cows for the second lactation could be explained by significantly ($p < 0.01$) higher milk yield associated with the AB phenotype. There were no significant association of α_{s1} -casein, β -casein and κ -casein types with yields and composition in the third lactation. β -lactoglobulin types were associated ($p < 0.05$) with protein content but not with fat percentage or milk, protein and fat yields. Figure 3 shows the protein content associated with different phenotypes of β -lactoglobulin for three lactations in Canadienne. During the third lactation, β -lactoglobulin AA phenotype was associated with higher protein in milk

Table 4. The f ratios from least squares analyses of variance for 305-D lactation yield and milk composition for three lactations in Canadienne

Source	df	Milk Yield	Fat Yield	Protein Yield	Fat (%)	Protein (%)
Lactation 1						
Herd	9	13.21**	12.33**	12.66**	0.84	2.80**
Age at calving	4	2.28	2.10	2.58	0.48	0.62
α_{s1} -Casein	1	0.04	0.15	0.18	0.05	0.77
β -Casein	4	1.39	0.91	1.19	0.48	1.53
κ -Casein	2	2.17	0.53	1.29	1.81	0.67
β -Lactoglobulin	2	1.18	0.68	0.42	2.62	0.95
Error	117					
Lactation 2						
Herd	8	12.22**	12.12**	14.92**	1.20	1.83
Age at calving	4	0.67	2.07	1.18	3.39*	0.87
α_{s1} -Casein	1	0.10	0.70	0.05	1.22	2.60
β -Casein	4	0.52	1.16	0.27	2.04	0.79
κ -Casein	2	5.01**	6.17**	4.92**	0.94	0.05
β -Lactoglobulin	2	0.76	0.97	0.26	0.26	1.01
Error	79					
Lactation 3						
Herd	8	6.38**	6.72**	6.21**	1.06	1.58
Age at calving	4	1.29	1.28	1.35	1.62	2.20
α_{s1} -Casein	1	1.39	2.08	2.04	0.14	0.48
β -Casein	4	0.59	0.72	0.35	1.29	1.23
κ -Casein	2	0.02	0.27	0.11	0.38	0.50
β -Lactoglobulin	2	0.28	0.50	0.08	1.62	4.89*
Error	41					

* $p < 0.05$, ** $p < 0.01$.**Figure 3.** Association between β -lactoglobulin phenotypes and protein content of milk for three lactations in Canadienne.

(3.79%) than either AB (3.54%) or BB (3.64%). Although not significantly different, the trend was also for higher

protein content to be associated with β -lactoglobulin AA for the first and second lactation. Results from the present study are similar to previous reports on Holstein which indicated that β -lactoglobulin AA milk contains higher protein percentage than the other two phenotypes, AB and BB (Cerbulis and Farrell, 1975; Feagan, 1979; Ng-Kwai-Hang et al., 1984, 1990). In those reports, protein contents are in the following decreasing order according to β -lactoglobulin types: AA > AB > BB. In the present study, the order is: AA > BB > AB during the first and third lactation. In the second lactation, the rankings were similar to those reported in the earlier reports above.

CONCLUSIONS

Frequency distributions of the different genetic variants of α_{s1} -casein β -casein, κ -casein and β -lactoglobulin are different for Brown Swiss, Canadienne and other breeds. After corrections are made for the effects of herd, age at calving, season of calving, any associations found between the phenotypes of a particular milk protein and lactational traits depend on the protein

system under consideration, lactation number, the traits in question and the breeds involved. In Brown Swiss no associations were found between phenotypes of α_{s1} -casein, β -casein and β -lactoglobulin and the lactational traits considered. κ -Casein types affected fat yield and protein yield during the first lactation, but not during the second and third lactation. For the Canadienne, there were no significant differences in yields and compositions for the different types of α_{s1} -casein, β -casein, κ -casein and β -lacto-globulin during the first lactation. In the second lactation κ -casein types were associated with yield parameters and in the third lactation, β -lactoglobulin types were associated with protein content of milk.

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