EFFECTS OF CASEIN HAPLOTYPES ON FIRST LACTATION MILK PRODUCTION TRAITS IN FINNISH AYRSHIRE COWS

T. Ikonen[1], H. Bovenhuis[2], M. Ojala[1] and O. Ruottinen[1]

[1] Department of Animal Science, University of Helsinki, P.O. Box 28, FIN-00014 Helsinki University, Finland, [2] Department of Animal Breeding, Wageningen Agricultural University, 6709 PG Wageningen, The Netherlands

SUMMARY

Effects of seven α_{s2}-β-κ-casein haplotypes and individual casein alleles on first lactation milk production traits were estimated for 16 973 Finnish Ayrshire cows by linear multiple regression analysis. The haplotypes AA_2A, AA_1E, AA_1B and DA_2B were clearly more frequent, whereas haplotypes AA_1A, AA_2B and AA_2E less frequent than expected assuming random combination of casein alleles. Haplotypes AA_1A, AA_1B and AA_1E were strongly associated with low milk and protein yields and high fat-%. Rare haplotype AA_2B had a favourable effect on milk and protein yields and AA_2A on protein yield. Haplotype AA_1B had a strong favourable effect on protein-%. The favourable effect of haplotype AA_2B on milk and protein yields and the unfavourable effect of that haplotype on fat-% and protein-% were more pronounced than the cumulative effects of individual α_{s2}-, β-, and κ-casein alleles on these traits. This indicates that there may be a QTL with alleles closely linked to haplotype AA_2B, which have a favourable effect mainly on milk yield, and an unfavourable effect on fat-% and protein-%.

Keywords: casein, haplotype, milk production, regression

INTRODUCTION

Association between bovine milk protein polymorphism and milk production traits has been estimated in several studies (e.g., Bovenhuis *et al.* 1992 and Ojala *et al.* 1997). However, due to differences for example in methods and models applied and linkage disequilibrium between the casein loci, there is not yet any uniform overview of direction and magnitude of effects of milk protein genotypes on milk production traits. It is possible that polymorphism at the casein loci is not solely causing the observed variation in milk production traits, but that these differences are partly or totally due to QTL linked to the casein loci. In this case it would be reasonable to estimate effects of casein genotypes within sires. However, if a mutation causing the differences in milk production traits exists within the casein complex, it is possible that this mutation is associated with a certain casein haplotype. It should be possible to reveal this by comparing if the effects of individual alleles add up to the effects of haplotypes constructed of these alleles. If that should be the case, it could be concluded that polymorphism at the casein loci is itself causing the observed differences in milk production traits.

The objective of this study was to estimate the effects of the α_{s2}-β-κ-casein haplotypes and the corresponding casein alleles on first lactation milk production traits in Finnish Ayrshire cows.

MATERIALS AND METHODS

A total of 18 686 Finnish Ayrshire (FAy) cows born during the years 1984-93 were genotyped for α_{s1}-, α_{s2}-, β- and κ-caseins (CN) and β-lactoglobulin using the IEF method (Erhardt 1989). The α_{s2}-β-κ-CN haplotypes were deduced by inheritance for 17 330 cows having at least nine paternal half-sibs. Haplotypes could be obtained for 17 068 cows. The 16 973 cows carrying two of the seven most common α_{s2}-β-κ-CN haplotypes were included in statistical analyses.

Effects of the α_{s2}-β-κ-CN haplotypes or the corresponding individual alleles on milk production traits were estimated assuming an animal model and a fixed model. Statistical significance of the regression coefficients could not, however, be tested when using the animal model. There were negligible differences in regression coefficients obtained assuming the previous models, which was due to the large data and reasonable number of daughters per sire. It was, therefore, justified to use the results obtained assuming the fixed model. The seven casein haplotype effects or allele effects were included simultaneously in the model as multiple regression coefficients to variables defining the number of each haplotype or allele a cow carried. The value of the last casein haplotype or allele could be concluded given the values of the other haplotypes or alleles. This dependency was solved by imposing a restriction $\Sigma b_o = 0$ on the regression coefficients as described by Østergård *et al.* (1989). The following model was used:

$$Y_{ijklmno} = \mu + yr_i + mo_j + do_k + h_l + s_m + \sum_n b_n x_{no} + \varepsilon_{ijklmno,}$$

where: $\mathbf{Y_{ijklmno}}$ = a milk production trait, μ = intercept, $\mathbf{yr_i}$ = fixed effect of i^{th} birth year class (i=1 - 5), $\mathbf{mo_j}$ = fixed effect of j^{th} birth month class (j=1 - 6), $\mathbf{do_k}$ = fixed effect of k^{th} days open class (k=1 - 6), $\mathbf{h_l}$ = fixed effect of l^{th} herd class (l=1 - 1 545), $\mathbf{s_m}$ = fixed effect of m^{th} sire (m=1-378), $\mathbf{b_n}$ = regression coefficient of a milk production trait on the number of copies of n^{th} casein haplotype (n=1 - 7) or n^{th} individual casein allele, $\mathbf{x_{no}}$ = 0, 1 or 2 depending on the number of n^{th} casein haplotype or allele for o^{th} cow, $\varepsilon_{\mathbf{ijklmno}}$ = random residual effect (0, σ_ε^2)

RESULTS AND DISCUSSION

The α_{s2}-β-κ-CN haplotypes AA_2A, AA_1E, AA_1B and DA_2B were more frequent, whereas AA_1A, AA_2B and AA_2E less frequent than expected assuming random segregation of casein alleles (Table 1). There are many potential reasons for disequilibrium between the casein loci, one being heavy use of few bulls with particular casein haplotypes. For example, occurrence of the rather frequent κ-CN E allele almost solely in combination with β-casein A_1 allele probably results from use of a popular sire in the 1970's (Velmala *et al.* 1995). Because of the low frequency of the haplotypes AA_2B, AA_2E and DA_2B, no cows were homozygous for them.

Haplotypes AA_1A, AA_1B and AA_1E were strongly associated with low milk and protein yields and high fat-% (Table 2). Haplotypes with β-casein A_2 allele had an opposite effect on previous traits. The effects of haplotype AA_2B on milk and protein yields, of AA_2A on protein yield, and of AA_2E on fat-% were statistically significant. Haplotype AA_1B had a clear favourable effect

on protein-%. Even though haplotypes AA_1A and AA_1E had a favourable effect on fat-%, they had an unfavourable effect on protein-%. When analysing the casein alleles individually, fat-% was affected by β-CN locus (A_1 favourable), whereas both β-CN locus (A_2 allele favourable) and κ-CN locus (B allele favourable) had a clear effect on protein-%.

Table 1. Observed and expected frequencies of α_{s2}-β-κ-CN haplotypes in FAy cows

α_{s2}-β-κ-CN haplotype	Observed N	Observed %	Expected %
AA_1A	5 319	15.7	31.1
AA_1B	2 146	6.3	3.9
AA_1E	9 805	28.9	15.5
AA_2A	15 605	46.0	30.1
AA_2B	221	0.7	3.8
AA_2E	596	1.8	14.9
DA_2B	254	0.7	0.0

Regression coefficient estimates for the individual α_{s2}-, β- and κ-CN alleles were added up in order to see if these cumulative allele effects were equal to the estimated haplotype effects. The effect of the rare haplotype AA_2B was clearly more favourable on milk yield, somewhat more favourable on fat and protein yields and considerably more unfavourable on fat and protein percentages when compared to the cumulative effects of α_{s2}-, β- and κ-CN alleles on these traits. This phenomenon may be explained by epistatic effects between the α_{s2}-, β- and κ-CN loci. Another explanation may be that beyond the casein loci there is a QTL with alleles closely linked to haplotype AA_2B, which have a favourable effect mainly on milk yield, and an negative effect on fat-% and protein-%. The effect of the AA_2B could also be due to chance.

Table 2. Regression coefficient estimates with statistical significance for α_{s2}-β-κ-CN haplotypes on first lactation milk production traits in FAy cows

α_{s2}-β-κ-CN haplotype	Milk yield, kg x=5 888	Fat yield, kg x=266	P yield, kg x=193 kg	Fat-% x=4.54	P-% x=3.29
AA_1A	-125 **	-1.1 ns.	-5.2 ***	0.07 **	-0.02 *
AA_1B	-239 ***	-3.6 †	-4.3 **	0.13 ***	0.07 ***
AA_1E	-69 *	1.2 ns.	-4.8 ***	0.07 ***	-0.04 ***
AA_2A	30 ns.	1.0 ns.	1.7 †	-0.02 ns.	0.01 †
AA_2B	185 †	1.2 ns.	7.0 *	-0.09 ns.	0.02 ns.
AA_2E	110 ns.	0.4 ns.	2.8 ns.	-0.08 *	-0.01 ns.
DA_2B	107 ns.	1.0 ns.	2.8 ns.	-0.08 ns.	-0.03 ns.

P = protein, x = mean, ***; p<0.001, **; p<0.01, *; p<0.05, †; p<0.10, ns.; non-significant

Associations between casein haplotypes and milk production traits have mostly been estimated assuming the granddaughter design (e.g., Lien *et al.* 1995, Velmala *et al.* 1995). In addition,

the entire chromosome six, where the casein loci are also located, has been screened for QTL affecting milk production traits using the previous method (e.g., Georges *et al.* 1995, Spelman *et al.* 1996). Then, associations between casein haplotypes, or other genetic markers, of sons of a heterozygous bull and for example sons' breeding values for the milk production traits have been estimated, which approach is quite different from that of this study. A significant favourable effect of β-κ-CN haplotype A_1E on fat-% and that of A_2A on milk yield was, however, observed also within one son group of a FAy grandsire (Velmala *et al.* 1995). In this study the rare haplotypes AA_2B and AA_2E, which did not occur in the study by Velmala *et al.* (1995), had a stronger positive effect on milk yield than AA_2A. The α_{s1}-β-κ-CN haplotype CA_5A was significantly associated with high milk and protein yields in Norwegian Cattle (Lien *et al.* 1995). A QTL having a favourable effect on milk yield and an unfavourable effect on fat-% and protein-% has been identified on chromosome six by Georges *et al.* (1995). Spelman *et al.* (1996) found a QTL affecting protein-% on chromosome six, which was, however, not located near the casein loci. According to Bovenhuis and Weller (1994), κ-CN locus had a direct effect on protein-% (B allele favourable), while fat-% was affected by β-casein locus and a closely linked QTL. Also in this study protein-% was clearly affected by κ-CN locus and fat-% by β-CN locus. In a sample of 916 Californian Holstein cows β-CN A_2 and κ-CN B alleles had a favourable effect on milk and protein yields only in genotype combinations with one another (Ojala *et al.* 1997), also suggesting a favourable association between β-κ-CN haplotype A_2B and milk and protein yields.

The α_{s2}-β-κ-CN haplotypes and possibly a QTL linked to the casein loci had a clear effect on two important traits in the breeding of the Finnish Ayrshire; protein yield and protein-%. However, using casein haplotypes as a selection criterion in order to breed for both previous traits does not seem reasonable. Selecting for AA_2B or AA_2A in order to increase protein yield would have a small effect on protein-%, but selecting for haplotype AA_1B in order to increase protein-% would probably result in a clear reduction in protein yield.

REFERENCES

Bovenhuis, H., van Arendonk, J.A.M. and Korver, S. (1992) *J. Dairy Sci.* **75**:2549-2559.

Bovenhuis, H. and Weller, J.I. (1994) *Genetics* **137**:267-280.

Erhardt, G. (1989) *J. Anim. Breed. Genet.* **106**:225-231.

Georges, M., Nielsen, D., Mackinnon, M., Mishra, A., Okimoto, R. et al. (1995) *Genetics* **139**:907-920.

Lien, S., Gomez-Raya, L., Steine, T., Fimland, E. and Rogne, S. (1995) *J. Dairy Sci.* **78**:2047-2056.

Ojala, M., Famula, T.R. and Medrano, J.F. (1997) *J. Dairy Sci.* **80**: in print.

Spelman, R., Coppieters, W., Karim, L., van Arendonk, J.A.M. and Bovenhuis, H. (1996) *Genetics* **144**:1799-1808.

Threadgill, D.W. and Womack, J.E. (1990) *Nucleic Acid. Res.* **18**:6935-6942

Velmala, R., Vilkki, J., Elo, K. and Mäki-Tanila, A. (1995) *Anim. Genet.* **26**:419-425.

Østergård, H., Kristensen, B. and Andersen, S. (1989) *Livest. Prod. Sci.* **22**:49-66.