

Possibilities of Genetic Improvement of Milk Coagulation Properties of Dairy Cows

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Academic dissertation

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Abstract

The objective of this thesis was to gather information on milk-coagulation properties to evaluate current means and needs to genetically improve them. This was done by collecting four separate data sets, which were used for estimating 1) genetic parameters for milk-coagulation properties, 2) milk protein polymorphism and its effect on milk-coagulation properties and milk-production traits, 3) occurrence of noncoagulating milk, 4) the association between coagulation properties of herd milk and cheese-making characteristics, and 5) differences in milk-coagulation properties between the main Finnish dairy breeds.

Based on the results presented in the thesis, genetic improvement of milk-coagulation properties would be a well-supported means to increase the efficiency of cheese production. Such an improvement would be possible by selection of animals directly for these traits or for properties associated with them.

Measurement of milk-coagulation properties is labourious and slow. Current possibilities to directly breed for these properties in the entire dairy population are thus limited. Because genetic improvement of milk-coagulation properties is most effectively achieved by direct selection for these properties, breeding values for milk-coagulation properties could, however, be estimated for an important group of dairy animals, e.g., bull dams and young AI-bulls.

Because none of the routinely recorded dairy traits strongly correlated with milk-coagulation properties in the present material, breeding for these traits seems not to markedly affect milk-coagulation properties. Breeding for high casein content and fat content may, however, have a favourable effect on cheese-making characteristics.

The κ -casein genotypes have a clear effect on coagulation properties, protein composition, and cheese-making properties of milk. Because of the rarity of the favourable B allele, and linkage disequilibrium between the κ -casein B allele and β -casein A₁ allele, which has an unfavourable effect on milk and protein yields, selection for the B allele in the entire Finnish dairy cattle would, however, be complicated.

The κ -casein E allele, which is rather common in the Finnish Ayrshire, is associated with poor milk-coagulation properties, low protein content, and, due to the linkage disequilibrium with the β -casein A₁ allele, also with low milk and protein yields. Selection against the κ -casein E allele would thus have a favourable effect on both milk-coagulation properties and important milk-production traits.

Noncoagulating milk, which was not affected by milk protein polymorphism or composition of milk, had an unfavourable effect on cheese-making characteristics. Noncoagulating milk was observed in the Finnish Ayrshire only. The environmental and genetic factors causing noncoagulation of milk should be established.

In conclusion, at present, information on breeding values for milk-coagulation properties and on κ -casein polymorphism for young AI-bulls and bull dams could be useful in genetically improving milk-coagulation properties in Finnish dairy cattle. Although such information would not serve as an official selection criterion of these animals, it could be made available for farmers who provide milk for cheese production, either at their own farms or at a commercial cheese plant.

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1 ORIGINAL ARTICLES

This thesis consists of an introduction and six papers, which deal with 1) variation in milk-coagulation properties, 2) association of milk-coagulation properties with cheese-making properties and milk-production traits, and 3) effect of milk protein polymorphism on milk-coagulation properties and milk-production traits. The following six papers included in the thesis are referred to by their Roman numerals:

- I** IKONEN, T., M. OJALA, and E-L. SYVÄOJA. 1997. Effects of composite casein and β -lactoglobulin genotypes on renneting properties and composition of bovine milk by assuming an animal model. *Agricultural and Food Science in Finland* 6:283-294.
- II** IKONEN, T., K. AHLFORS, R. KEMPE, M. OJALA, and O. RUOTTINEN. 1999. Genetic parameters for the milk coagulation properties and prevalence of noncoagulating milk in Finnish dairy cows. *Journal of Dairy Science* 82:205-214.
- III** IKONEN, T., O. RUOTTINEN, E-L. SYVÄOJA, K. SAARINEN, E. PAHKALA, and M. OJALA. 1999. Effect of milk coagulation properties of herd bulk milks on yield and composition of Emmental cheese. *Agricultural and Food Science in Finland* 8:411-422.
- IV** IKONEN, T., O. RUOTTINEN, G. ERHARDT, and M. OJALA. 1996. Allele frequencies of the major milk proteins in the Finnish Ayrshire and detection of a new κ -casein variant. *Animal Genetics* 27:179-181.
- V** IKONEN, T., M. OJALA, and O. RUOTTINEN. 1999. Associations between milk protein polymorphism and first lactation milk production traits in Finnish Ayrshire cows. *Journal of Dairy Science* 82:1026-1033.
- VI** IKONEN, T., H. BOVENHUIS, M. OJALA, O. RUOTTINEN, and M. GEORGES. 2000. Associations between casein haplotypes and first lactation milk production traits in Finnish Ayrshire cows. Accepted for publication in *Journal of Dairy Science*.

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Contribution of the author to papers I to VI:

- I. The author participated in planning of the study and in quantification of milk proteins, genotyped the cows, determined milk coagulation properties, conducted the statistical analyses, interpreted the results, and was the main author of paper I.
- II. The author participated in planning of the study and in the statistical analyses, and was the main author of paper II.
- III. The author participated in planning and carrying out the experiment, conducted the statistical analyses, interpreted the results, and was the main author of paper III.
- IV-VI. The author participated in the planning of the study and in milk protein genotyping, carried out the statistical analyses, interpreted the results, and was the main author of papers IV-VI.

2 INTRODUCTION

2.1 Cheese production in Finland

In Finland, the dairy industry is the third most important branch of the food industry (Finnish Food and Drink Industries' Federation 1998). Within the dairy industry, cheese is a product of major importance; about 40% of the milk produced in Finland is used for cheese production (Agrifacts 1998). About 30% of the net sales of the Valio Group, which consists of the biggest Finnish dairy company Valio Ltd. (Helsinki, Finland) and its subsidiaries, comes from cheese production (Valio Ltd. 1999). Further, during the 1990s, domestic consumption of cheese has increased about 20% (Agrifacts 1998).

Finland joined the European Union (EU) in 1995, which caused remarkable changes in the Finnish dairy industry, and in the foreign trade in Finnish dairy products. Because of EU membership, Finnish dairy products have to compete with those of the other EU states in the EU internal market. Since the year 1995, import of cheese from the other EU states has thus markedly increased in Finland (Finnish Food and Drink Industries' Federation 1998). In addition, because of various structural changes in the Finnish dairy industry during previous years, competition between local dairies has tightened.

Because of the present, rather challenging situation of the Finnish cheese industry, it is important that the profitability of domestic cheese production is maintained and improved. This could be accomplished through identification and improvement of the factors that affect efficiency of cheese production: cheese yield, dry-matter cheese yield, and composition of cheese and cheese whey.

2.2 Milk coagulation process and cheese-making

Enzymatic coagulation of milk is a process of three overlapping steps (BROWN and ERNSTROM 1988), which can be described with a diagram produced by a milk-coagulation meter (Figure 1). During the primary, enzymatic phase (R in Figure 1), chymosin, which is the clotting enzyme extracted from calf abomasum, splits κ -casein at the Phe₁₀₅-Met₁₀₆ bond into para- κ -casein and a macropeptide. Because of this splitting of κ -casein, casein begins to aggregate. This second, non-enzymatic phase of milk coagulation begins before all of the κ -casein has been split.

During the third step of milk coagulation, aggregated casein micelles form a more or less firm gel structure. Curd-firming time, K_{20} , describes the time needed until the curd is firm enough to be cut (= the width of the diagram (Figure 1) is 20 mm), and curd firmness, E_{30} , describes the firmness of the curd 30 min after addition of the clotting enzyme. These milk-coagulation properties (MCP) are measured for 30 min or more, because in cheese-making for most cheese types, the curd is cut about 30 min after addition of the clotting enzyme to the milk.

MCP and composition of milk have a rather clear effect on cheese-making properties. Milk that begins to aggregate soon after addition of the enzyme, and forms a firm curd within a reasonable time is expected to produce higher dry-matter cheese yields than does milk with unfavourable coagulation properties (RIDDELL-LAWRENCE and HICKS 1989, BYNUM and OLSON 1982, ALEANDRI et al. 1989). This occurs because milk that coagulates quickly is able to entrap more casein and fat into the coagulum before it is cut than does slowly coagulating milk. Casein and fat constitute about 90% of the solids in cheese, so the amount of casein and fat lost in the cheese whey has a substantial effect on the efficiency of cheese-making (JOHNSON 1988, POLITIS and NG-KWAI-HANG 1988a, 1988b, and LAWRENCE et

al. 1993). Because the possibility to vary the cutting point is limited in commercial large scale cheese production, it is important that the curds are firm enough to allow cutting at the usual cutting time.

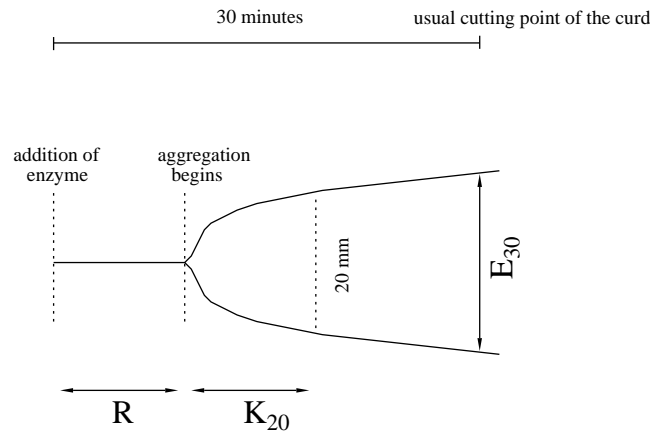


Figure 1: Diagram produced by a milk-coagulation meter, and the three milk-coagulation parameters calculated from the diagram: R, milk coagulation time; K_{20} , curd-firming time; E_{30} , curd firmness 30 min after addition of the clotting enzyme.

2.3 Established knowledge of MCP in Finnish dairy cattle

2.3.1 Phenotypic variation in MCP

Because MCP are not determined routinely for Finnish dairy cows in milk recording, no official statistics exist as to phenotypic variation or the trend in these characteristics. However, based on the observations made in Finnish commercial cheese dairies over the past 20 to 30 years, MCP may have been deteriorating.

In the 1980s, variation in MCP in the Finnish dairy breeds, Finnish Ayrshire (FAy), Finnish Friesian (FFr), and Finncattle (Fc), and factors causing the variation were examined in a few studies (TERVALA et al. 1983, LINDSTRÖM et al. 1984, TERVALA et al. 1985, TERVALA and ANTILA 1985, and AALTONEN and ANTILA 1987). The data sets in these studies were rather small; the number of milk samples ranged from less than hundred to about 700.

Based on the studies of TERVALA et al. (1983, 1985) and LINDSTRÖM et al. (1984), wide variation exists in MCP between cows. About 13% of the cows produced milk that received no value for curd-firming time (TERVALA et al. 1983, 1985). Further, 2% of the cows in TERVALA et al. (1983), and 4% of the FAy cows in TERVALA et al. (1985) produced non-coagulating milk. According to TERVALA and ANTILA (1985), the enzymatic phase of the milk coagulation process (Figure 1) proceeded normally in the noncoagulating milk, but no curd was formed. Variation in protein or mineral composition of milk failed to explain non-coagulation of milk (TERVALA and ANTILA 1985). Because only one noncoagulating milk sample from one cow was studied, it was impossible to draw general conclusions regarding the factors causing it.

2.3.2 Genetic variation in MCP

Even though the heritability estimates of MCP in LINDSTRÖM et al. (1984) and in TERVALA et al. (1985) were based on small samples and thus had large standard errors, they indicate that a moderate part of the variation in these properties is genetic in nature.

Genetic correlations between MCP and milk-production traits were estimated for milk coagulation time only (LINDSTRÖM et al. 1984). Short coagulation time was associated with low pH, high fat content and high protein content of milk.

In addition to the above studies, genetic parameters for MCP have been estimated in one study only (OLOFFS et al. 1992). The heritability estimates for MCP reported by OLOFFS et al. (1992) were somewhat higher than those in LINDSTRÖM et al. (1984) and TERVALA et al. (1985). Genetic correlations between MCP and milk-production traits could not be reliably estimated in OLOFFS et al. (1992).

In the above studies, the number of cows ranged from a few hundred to less than 2000, and the genetic parameters were estimated by use of a sire model under the least squares procedure, which probably impeded reliable estimation of the above parameters.

2.3.3 Genetic factors affecting MCP

Breed. In the FAY, the proportion of cows producing milk that received no value for coagulation time or curd-firming time or both was higher than in the FFR and Fc (TERVALA et al. 1985). Differences in the MCP between the above breeds were, however, statistically non-significant (TERVALA et al. 1983 and 1985).

Milk protein polymorphism. κ -casein polymorphism was one clear reason for the variation in MCP (TERVALA et al. 1983, 1985, and AALTONEN and ANTILA 1987). It is possible that the favourable effect of the κ -casein B allele on these properties was partly due to the high κ -casein content in milk (AALTONEN and ANTILA 1987).

Differences in coagulation properties (TERVALA et al. 1985 and AALTONEN and ANTILA 1987) and composition of milk (AALTONEN and ANTILA 1987) between the milk protein genotypes were tested by comparing the uncorrected means of these traits for the individual genotypes. Simultaneous estimation of the effect of milk protein polymorphism and, for example, of the effect of κ -casein content of milk on coagulation properties was impossible in AALTONEN and ANTILA (1987). In the above studies, estimation of the underlying effect of κ -casein polymorphism on MCP was thus impossible.

Several other studies have, however, also shown that the κ -casein B allele has a more favourable effect on coagulation or cheese-making properties, and on protein, casein, and κ -casein contents of milk than the A allele (e.g. SCHAAR 1984, SCHAAR et al. 1985, MARZIALI and NG-KWAI-HANG 1986, PAGNACCO and CAROLI 1987, DAVOLI et al. 1990, RAHALI and MÉNARD 1991, VAN DEN BERG et al. 1992, WALSH et al. 1995, and LODES et al. 1996a, 1996b, 1997). In these studies, the effect of milk protein polymorphism on MCP was estimated without taking into account the relationships between animals. It is, however, possible that the animals sharing the same milk protein alleles were closely related. Information on relationships between animals is thus necessary for adjusting the effects of the milk protein genotypes for polygenic effects.

The κ -casein E allele was detected only after an improved isoelectric focusing method was developed in 1989 by ERHARDT (1989). In TERVALA et al. (1983 and 1985) and in AALTONEN and ANTILA (1987), estimation of the frequency of the κ -casein E allele, and of the effect of this allele on MCP was thus impossible.

No clear results were obtained for the effects of genotypes of the other major milk proteins (α_{s1} - and β -caseins, and β -lactoglobulin) on MCP in TERVALA et al. (1983 and 1985) or in AALTONEN and ANTILA (1987). In the literature, the β -lactoglobulin B allele has, however, been shown to have a favourable effect on casein content (MCLEAN et al. 1984, NG-KWAI-HANG et al. 1986), casein number (NG-KWAI-HANG et al. 1986), and fat content (ALEANDRI et al. 1990). It is thus possible that β -lactoglobulin polymorphism is of indirect importance for cheese-making characteristics through its effect on composition of milk.

2.3.4 Other factors affecting MCP

Environmental factors, such as season, parity, lactation stage, and feeding (SCHAAR 1984, KREUZER et al. 1996, DAVOLI et al. 1990, OKIGBO et al. 1985), and various technological factors effective before or during cheese-making (e.g., time and temperature of milk storage, heat treatment, homogenisation and standardisation of milk, type and concentration of the clotting enzyme, addition of calcium, and cooking time and temperature) may also affect MCP (ALEANDRI et al. 1989, and RIDDELL-LAWRENCE and HICKS 1989). In this thesis, environmental factors affecting MCP are studied briefly, whereas technological factors are not addressed.

2.4 Further information needed for genetic improvement of MCP

The variation in MCP and the relatively high proportion of cows producing poorly coagulating or noncoagulating milk (TERVALA et al. 1983 and 1985, LINDSTRÖM et al. 1984) indicate that improvement of MCP in the Finnish dairy cattle is both possible and necessary. MCP could be improved by varying the environmental or technological factors that affect MCP or by breeding for these properties. The advantage of breeding over that of changing various environmental factors is that genetic improvement is permanent and cumulative.

In principal, genetic improvement of MCP is possible by selecting the breeding animals for these properties or for dairy traits that are strongly correlated with MCP. Different kinds of information are necessary to evaluate the possibilities of direct and indirect breeding for these properties.

2.4.1 Selection for MCP

Genetic parameters. The heritability estimates in LINDSTRÖM et al. (1984) and TERVALA et al. (1985) indicate that direct selection for MCP is possible. Reliable heritability estimates for MCP are thus needed for accurate genetic evaluation of animals and for accurate estimation of the milk protein genotype effects on MCP.

Repeatability estimates for MCP are needed to evaluate the number of measurements for MCP needed for genetic evaluation of these properties. Estimates of the genetic associations between MCP and important dairy traits (milk-production, fertility, and health traits) are needed to evaluate how breeding for MCP would affect these traits.

Reliable estimation of the above genetic parameters for MCP should be possible by collecting data that are large enough, and by utilizing an animal model in the statistical analyses. Because an animal model accounts for the known relationships between animals, it provides more accurate estimates for the genetic (co)variances for MCP than does the least squares procedure.

Noncoagulating milk. Noncoagulation of milk, which could not be explained by protein or

mineral composition of milk, was observed in two samples collected in the 1980s (TERVALA et al. 1983 and 1985). It is important to establish the current prevalence of this phenomenon in Finnish dairy cattle, and to identify the environmental and genetic factors causing it.

Variation in MCP between herds and Finnish dairy breeds. MCP are likely to vary greatly between the Finnish dairy cows, with no information existing on variation in these traits between dairy herds, and on the effect of this variation on cheese-making conditions at cheese plants. For a cheese dairy, information on the variation in MCP between herds may be more important than on variation between animals. In addition, it is important to reliably estimate the difference in MCP between Finnish dairy breeds.

2.4.2 Selection for associated characteristics

κ -casein polymorphism. The favourable effect of the κ -casein B allele on MCP reported in the literature indicates that these properties could be improved by increasing frequency of the B allele in dairy cattle.

Before information on κ -casein polymorphism can be used in selection, however, reliable estimates are needed for the occurrence of the known κ -casein alleles, and for their effects on MCP. Further, estimates for the effect of κ -casein polymorphism on milk-production, fertility, and health traits are needed to evaluate how selection for certain κ -casein alleles to improve MCP would affect these traits.

Because the alleles of the α_{s1} -, β -, and κ -caseins may be in linkage disequilibrium due to the close location of the casein loci on chromosome six (FERRETTI et al. 1990, THREADGILL and WOMACK 1990), polymorphism of the other caseins has to be taken into account in estimating the effect of κ -casein polymorphism on the above traits. Further, because β -lactoglobulin polymorphism can be important in cheese-making, its effect on milk-production, fertility, and health traits needs to be estimated, as well.

In the literature, results concerning the effect of milk protein polymorphism on milk-production traits are inconsistent (GRAML et al. 1986, LIN et al. 1986, GONYON et al. 1987, HAENLEIN et al. 1987, LIN et al. 1989, ALEANDRI et al. 1990, NG-KWAI-HANG et al. 1990, BOVENHUIS et al. 1992, MAO et al. 1992, BOVENHUIS and WELLER 1994, FAMULA and MEDRANO 1994, RON et al. 1994, SABOUR et al. 1996, and OJALA et al. 1997), and those on health (ATROSHI et al. 1982), and fertility (HARGROVE et al. 1980, and LIN et al. 1987) traits rare.

In these studies, size and structure of the data sets (linkage disequilibrium in the alleles of the casein loci), dependent variables, and statistical procedures used differ, which is one probable explanation for the conflicting results. For example, an animal model, which should be used to estimate effects of single genes (KENNEDY et al. 1992), was utilized in four of these studies only.

Associated dairy traits. Estimates of genetic associations between MCP and the dairy traits routinely measured for dairy cows are needed to estimate whether MCP can be improved by selecting for or against associated dairy traits.

3 THE OBJECTIVE OF THE STUDY

The results from the studies performed during the 1980s in Finland (TERVALA et al. 1983, 1985, LINDSTRÖM et al. 1984, and AALTONEN and ANTILA 1987) indicate that both the need and potential exist for genetic improvement of MCP in Finnish dairy cattle. The objective of this thesis is to gain further information on MCP to evaluate current possibilities and alternatives for genetically improving them.

The main subjects discussed in this thesis are: 1) genetic parameters for MCP, 2) milk protein polymorphism and its effect on MCP and milk-production traits, 3) occurrence of noncoagulating milk, 4) the association between coagulation properties of herd milk and cheese-making characteristics, and 5) differences in MCP between the main Finnish dairy breeds.

Heritability of MCP, and the effect of milk protein polymorphism on MCP were estimated in two studies (I, II). In I, reasons for the effect of milk protein polymorphism on MCP were also studied by measurement of milk samples for various composition characteristics.

Genetic correlations between MCP and milk-production traits, and breeding values for these traits were estimated in II.

Occurrence of noncoagulating milk, and the difference in MCP between FAy and FFr cows were evaluated in I and II.

Because wide variation existed in MCP among the herds in II, the effect of this variation on cheese-making characteristics was evaluated in III.

Milk protein polymorphism and its effect on first-lactation milk-production traits were estimated in IV to VI. A large data set was collected to estimate the effect of milk protein polymorphism on these traits by various means (using individual genotypes, composite genotypes, and casein haplotypes), and by use of appropriate statistical models.

4 MATERIALS AND METHODS

4.1 Data sets and characteristics studied

For this thesis, four separate data sets were collected during the years 1990 to 1997 (Table 1). In I to III, milk samples of both FAy and FFr cows were collected, whereas the data in IV to VI included FAy cows only.

The type and number of milk samples, and the chemical analyses carried out differed between the data sets in I to VI. The characteristics studied can, however, be classified into three groups according to their nature and utilisation:

1. Coagulation, composition, and cheese-making properties of milk (I – III)
2. Milk protein polymorphism (I – VI)
3. First-lactation milk-production traits (V – VI).

4.1.1 Coagulation, composition, and cheese-making properties of milk

MCP. MCP (Figure 1) were determined with a Formagraph (Foss Electric A/S, Hillerød, Denmark) at Valio Ltd. (I, III), or at the Agricultural Research Centre at Jokioinen, Finland (II). In I and II, daily milk samples of individual cows were used for measuring the coagulation properties. The cows in II were sampled once during lactation, and those in I two or three times. In III, MCP were determined for the bulk milk samples of 30 candidate herds, and additionally for the bulk milk samples of the eight herds that provided milk for the cheese-making experiment.

Composition of milk. All milk samples in I to III were measured for fat content and protein content (Milko Scan 605, Foss Electric A/S), somatic cell count (Fossomatic cell counter, Foss Electric A/S), and pH at the laboratories involved in milk recording, or at Valio Ltd.

Detailed protein composition of milk (contents of casein, whey protein, α_{s1} -, α_{s2} -, β -, and κ -caseins, α -lactalbumin, and β -lactoglobulin) was determined for the milk samples in I, and for the bulk milk samples in III. In addition, ash, calcium, and phosphorus contents were determined for the bulk milks in III. These characteristics were measured at Valio Ltd., and descriptions of the methods used to measure them are presented in I and III.

Cheese-making experiment. The cheese-making experiment was carried out at a pilot-scale cheese plant (III). Of the eight herds that provided milk for the experiment, four herds produced moderately coagulating milk (A herds), and four herds extremely poorly coagulating milk (B herds). The milks of the two herd groups, which were from two to six milkings, were standardised to the same fat content. Two cheeses were made of the well-coagulating bulk milk, and two cheeses of the poorly coagulating milk. About 750 l of milk was used to produce one cheese.

The cheeses were made according to a standard procedure used in commercial production of Emmental cheese. Fresh and ripened cheeses were analysed for contents of dry-matter, fat, protein, ash, calcium, and phosphorus and for moisture in the non-fat substance (for fresh cheese), and cheese whey for contents of dry-matter, fat, protein, casein and caseinomacropptide. Ripened cheeses were assessed for interior appearance, texture, and taste. A description of the above traits and of the methods used to determine them appears in III.

Other information. Information on parity, lactation stage, season, breed, herd, and pedigree

Table 1: Basic information of data sets of papers I to VI

Paper	No. of herds	Breed of cows	No. of cows	No. of samples ¹	Year of coll. ²	Location ³	Main objectives of the paper
I	2	FAy ⁴ FFr ⁵	59 55	174 155	1990 - 91	Helsinki and Siuntio	Associations of milk protein genotypes with coagulation properties and protein composition of milk
II	51	FAy FFr	789 86	789 86	1995	Ad. ⁶ of Southern Finland	Genetic parameters for milk coagulation properties, and prevalence of noncoagulating milk
III	8-30	FAy FFr	47 65	4-112 ⁷	1997	Alueosuuskunta Promilk ⁸	Effect of milk coagulation properties of herd bulk milk on yield and composition of Emmental cheese
IV	1688	FAy	20 990	20 990	1994 - 95	Alueosuuskunta Promilk ⁹	Allele frequencies of the major milk proteins
V	1548 ¹⁰	"	18 686	18 686	"	"	Effect of milk protein genotypes on first lactation milk production traits
VI	1545 ¹⁰	"	16 973	16 973	"	"	Effect of casein haplotypes on first lactation milk production traits

¹Milk samples from individual cows.

²Year(s) data were collected.

³Area from which data were collected.

⁴Finnish Ayrshire.

⁵Finnish Friesian.

⁶Administrative district.

⁷Samples of individual cows or herd bulk milk samples.

⁸Herds within one milk collection route of the dairy co-operative Alueosuuskunta Promilk in Lapinlahti.

⁹Herds that provide milk for Alueosuuskunta Promilk, and are located within an area of 35 communes in central and eastern Finland.

¹⁰A sub-sample of the data in IV.

of the cows in I to III required in the statistical analyses was obtained from the Agricultural Data Processing Centre, Vantaa, Finland.

4.1.2 Milk protein polymorphism

For each cow in I to VI, genotypes of α_{s1} -, β -, and κ -caseins, and β -lactoglobulin (the term genotype is used instead of phenotype) were determined in polyacrylamide gels as described by ERHARDT (1989) at the Finnish Animal Breeding Association, Vantaa, Finland. The milk samples used for genotyping were taken from each cow before milking.

4.1.3 First-lactation milk-production traits

Records for the first-lactation milk-production traits (milk, fat, and protein yields, and fat and protein contents), and information on various factors likely to affect them (herd, month and year of birth, calving and insemination, and pedigree) were obtained from the milk recording data base from the Agricultural Data Processing Centre.

4.2 Statistical analyses

4.2.1 Genetic parameters

The (co)variance components for the random effects in the statistical models, which were used to estimate 1) heritability or repeatability, or both, of MCP (I, II), and first-lactation milk-production traits (V), 2) genetic correlations between the above traits (II) were estimated from the data by use of an animal model and the REML VCE -package (GROENEVELD 1996).

4.2.2 Cheese-making experiment

Statistical significance of the differences in composition, coagulation, and cheese-making characteristics of milk between the two herd groups was tested by one-way analyses of variance, and the *F* test (III).

4.2.3 Milk protein polymorphism

Allele frequencies of the caseins and β -lactoglobulin were calculated from the corresponding genotypes by the gene-counting method (I to VI). The β - κ -casein haplotypes were deduced by inheritance for the FAy cows in V that had at least nine paternal half-sibs (VI). A description of the program used to deduce the haplotypes is presented in VI.

Expected genotype frequencies of the caseins, composite β - κ -casein and β -lactoglobulin, and expected frequencies of the β - κ -casein haplotypes were calculated by multiplication of the allele frequencies of the corresponding proteins.

4.2.4 Effect of milk protein polymorphism

Effect of milk protein polymorphism on coagulation properties and composition of milk (I, II), and on first-lactation milk-production traits (V, VI) was estimated by an animal model. Statistical significance of the milk protein genotype (I, II, V) or casein haplotype (VI) effects were tested with the *F* test provided by the PEST package (GROENEVELD 1990).

Effects of the β -casein and κ -casein genotypes were estimated by means of composite β - κ -casein genotypes (I, II, V) and β - κ -casein haplotypes (VI). Because the cows in these

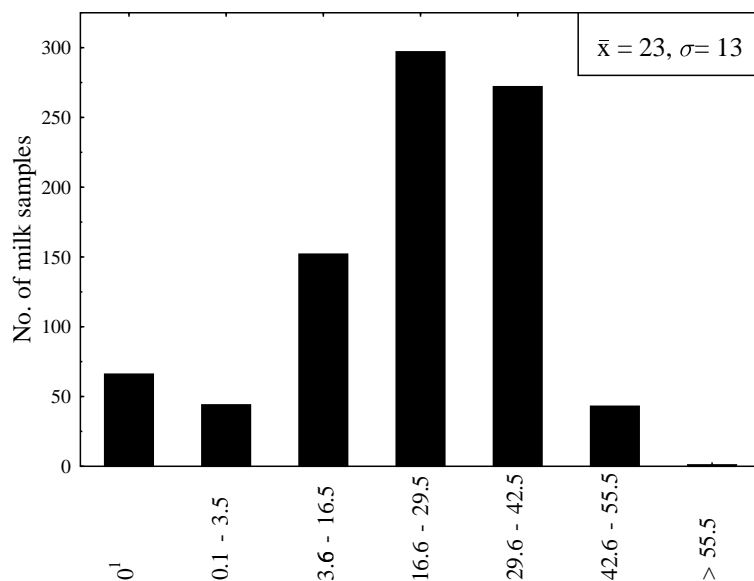
studies were almost monomorphic for the α_{s1} -casein, these caseins were excluded from the statistical analyses. Effects of the β -lactoglobulin genotypes were estimated separately from the casein genotype or haplotype effects (I, II, V, VI).

Composite casein genotypes and casein haplotypes were used in the statistical analyses, because the casein loci, which are located at chromosome six within a region of 200 to 250 kb (FERRETTI et al. 1990, THREADGILL and WOMACK 1990, and RIJNKELS et al. 1997), are tightly linked. Certain combinations of the alleles at the casein loci may thus appear either more often or more rarely than expected when a random combination of casein alleles is assumed. The data structure may thus be unbalanced, and the effects of the casein genotypes may depend on each another.

5 RESULTS

5.1 Phenotypic variation in MCP

MCP varied considerably among the cows; the coefficient of variation for these properties ranged from 33 to 57% (I, II). In addition, 23 and 33% of the milk samples of the FAY cows (I, II), and 8% of those of the FFr cows (I) received no values for curd-firming time (K_{20}), meaning the curds of these milk samples were too soft to be cut at the usual cutting point. Further, 5% and 8% of the samples of the FAY cows did not coagulate in 30 min (I, II). For these samples, no values for milk coagulation time (R) and curd-firming time were produced and the value for curd firmness (E_{30}) was 0.0. Curd firmness was thus the most informative and the most important characteristic. In I, the distribution of values for curd firmness was skewed towards the most unfavourable values (Figure 2).



Minimum and maximum values for curd firmness (E_{30}) in the seven classes

Figure 2: Distribution of values for curd firmness, data (n=875) adapted from Paper II.
¹Noncoagulating milk samples.

5.2 Environmental factors affecting MCP

5.2.1 Parity and lactation stage

MCP were at their best at the beginning and at the end of lactation (II). Part of this variation was caused by the changes in milk yield, protein content, fat content, and somatic cell count during lactation. Parity had no statistically significant effect on MCP (II).

5.2.2 Herd

MCP varied considerably among the herds (II, III). Four herds in III produced noncoagulating milk. In II, milk protein polymorphism explained a minor part of the variation in MCP among the herds. Environmental factors, such as feeding and management, may have been one explanation for the variation. In III, the κ -casein B allele was more frequent in the four herds with the most favourable MCP (19%) than in those with the poorest (4%).

5.3 Genetic factors affecting MCP

5.3.1 Breed

MCP were better for the FFr than for the FAy cows (I, II). Furthermore, the proportion of FFr cows was higher (86%) in the herds producing well-coagulating milk than in those producing poorly coagulating milk (18%) (III). None of the FFr cows produced noncoagulating milk, which explained, however, only part of the differences in MCP between the breeds.

5.3.2 Milk protein polymorphism

In general, the β - κ -casein genotypes including the κ -casein B allele were associated with the most favourable MCP, and those with the E allele with the poorest (I, II). In addition, the breeding value estimates for curd firmness were highest for the cows carrying the κ -casein AB genotype, and lowest for those carrying the AA, AE or EE genotype (II). The favourable effect of the κ -casein B allele on MCP was partly due to high κ -casein content in milk (I).

The β -casein genotypes had no clear effect on MCP (I, II). Further, although the β -lactoglobulin B allele was associated with high casein content and casein number (I), the effect of β -lactoglobulin polymorphism on MCP was negligible.

Milk protein polymorphism explained a moderate proportion (3 to 16% in I, and 20 to 24% in II) of the additive genetic variation in MCP. Based on the effects of the β - κ -casein and β -lactoglobulin genotypes on MCP, a major proportion of this contribution was due to κ -casein polymorphism.

The casein or β -lactoglobulin alleles were not clearly associated with noncoagulation of milk (I, II). It is, however, possible that other genetic factors cause this phenomenon; some of the cows producing noncoagulating milk were sired by seven bulls, which constituted three families based on their mutual sire or grandsire or both.

5.4 Genetic parameters

5.4.1 Heritabilities

MCP are well-inherited and repeatable traits; the heritability estimates ranged from 0.22 to 0.62 for milk coagulation time, and from 0.40 to 0.57 for curd firmness (I, II). The heritabilities estimated from the small amount of data (I) were somewhat higher than those estimated from the data in II. Repeatability estimates for MCP ranged from 0.57 to 0.71 (I).

5.4.2 Genetic correlations

MCP were highly correlated with one another (II), which was logical, because they describe the partially overlapping steps of the milk-coagulation process.

Except for protein content and pH, no reliable estimates for the genetic correlations between MCP and milk-production traits were achieved (II). The unfavourable genetic correlation between high protein content and milk coagulation ability was rather unexpected. High pH of milk also correlated unfavourably with MCP.

5.5 Associations between MCP and cheese-making properties

The eight herds that provided milk for the cheese-making experiment differed markedly in coagulation properties and protein composition of their milk (III). The four herds that produced poorly coagulating milk before the experiment provided noncoagulating milk (B milk) for the experiment. The herds in the other group provided well-coagulating bulk milk (A milk). Concentrations of κ -casein, α -lactalbumin, and β -lactoglobulin were higher in A milk than in B milk.

MCP of the herd bulk milks had a clear effect on some of the cheese-making characteristics and thereby on the efficiency of cheese production. Dry-matter content and contents of calcium and phosphorus were higher ($P < 0.10$ to $P < 0.01$) for the cheeses made of A milk (A cheeses) than for those made of B (B cheeses), because less fat ($P < 0.05$) and casein ($P < 0.10$) were lost in the cheese whey of A cheeses. In addition, the ratio of moisture to non-fat substance (MNFS) was lower ($P < 0.05$) for A than for B cheeses. A high MNFS in cheese is likely to be associated with higher rate of casein proteolysis in cheese and thus with a more unfavourable quality for that cheese than is a low MNFS (Politis and Ng-Kwai-Hang 1988a).

5.6 Milk protein polymorphism

For the FAy, frequencies of milk protein genotypes and alleles could be estimated from samples in I (59 cows), II (789), III (47), and IV to VI (16 973 to 20 990). For the FFr, these frequencies were estimated from three small samples in I (55 cows), II (86), and III (65). Because milk protein genotype frequencies for the FAy that were estimated from the samples in I to III agreed well with those estimated from the large sample (V), it is possible that the estimates of milk protein polymorphism in the FFr samples (I to III) represent well those in the FFr population. In addition, the κ -casein and β -lactoglobulin allele frequencies observed in the FFr samples (I to III) agreed well with those presented for 42 FFr bulls in VELMALA et al. (1993).

5.6.1 Allele frequencies

In the FAy samples, α_{s1} -casein was monomorphic, and the β -casein A₁ and A₂ alleles were equally frequent (Table 2). The most frequent κ -casein allele was A, with the E allele also rather common. Frequency of the κ -casein B allele was low, and the β -lactoglobulin B allele clearly more frequent than the A allele.

The FAy and FFr cows differed in allele frequencies of each milk protein. In the FFr, the α_{s1} -casein C allele, the β -casein A₃ and B alleles, the κ -casein B allele, and the β -lactoglobulin A allele were more frequent, and the κ -casein E and the β -lactoglobulin B alleles less frequent than in the FAy (Table 2).

5.6.2 β - κ -casein genotype and haplotype frequencies

Because of the small numbers of samples from FFr cows, frequencies of composite casein genotypes and casein haplotypes are presented for the FAy only. The most frequent β - κ -

Table 2: Frequencies (%) of the α_{s1} -casein, β -casein, κ -casein, and β -lactoglobulin alleles in a sample of Finnish Ayrshire (FAy) cows in IV (n=20 990), and in three separate samples of Finnish Friesian (FFr) cows in I (n=55), II (n=86), and III (n=65).

Milk protein	Allele	FAy	FFr
α_{s1} -casein	B	100	78 – 87
	C		13 – 22
β -casein	A ₁	51	31 – 51
	A ₂	49	47 – 65
	A ₃		0 – 1
	B		0 – 3
κ -casein	A	61	71 – 81
	B	8	14 – 17
	E	31	6 – 13
β -lactoglobulin	A	28	42 – 56
	B	72	45 – 58

casein genotypes, A₁A₂AE and A₂A₂AA, constituted more than half of the data in V (Figure 3). Combinations of β -casein genotypes A₁A₂ and A₂A₂, and κ -casein genotypes BB, BE, and EE were rare or non-existent.

Expected frequencies of the composite β - κ -casein genotypes differed from those observed (Figure 3). Among the cows carrying β -casein A₁A₁ genotype, κ -casein BB, BE, and EE genotypes were more frequent, and genotype AA less frequent than expected, if a random combination of casein alleles is assumed. Further, practically all cows carrying the κ -casein EE genotype carried the β -casein A₁A₁ genotype, as well.

Within the β -casein A₁A₂ genotype, the κ -casein AE genotype was more frequent, and genotypes AA, BB, BE, and EE less frequent than expected. Among the cows carrying the A₂A₂ genotype, nearly all cows carried the κ -casein AA genotype, even though κ -casein genotypes AA and AE had been expected to be equally frequent.

These differences in expected and observed frequencies of the β - κ -casein genotypes were reflected also in the β - κ -casein haplotypes (VI). Haplotypes A₁B, A₁E, and A₂A were more frequent, and haplotypes A₁A, A₂B, and A₂E less frequent than expected. Because of linkage disequilibrium in the β - and κ -casein loci, the data sets were unbalanced regarding these casein genotype frequencies (I to VI).

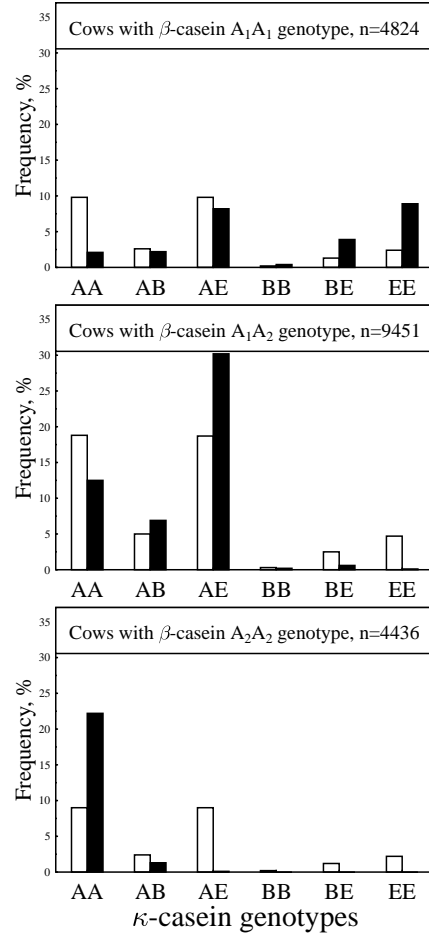


Figure 3: Expected (white bars) and observed (black bars) frequencies of κ -casein AA, AB, AE, BB, BE, and EE genotypes within β -casein genotypes A_1A_1 , A_1A_2 , and A_2A_2 . Data (18 742 FAY cows) adapted from Paper V, Table 1.

5.7 Effect of milk protein polymorphism on first-lactation milk-production traits

5.7.1 β - κ -casein genotypes

The rare β - κ -casein genotype A_2A_2AB , and the most frequent genotypes A_2A_2AA and A_1A_2AE were associated with the highest milk and protein yields, and lowest fat content of milk (V). The β - κ -casein genotypes including the β -casein A_1 allele were associated with low milk and protein yields and high fat content. These traits were thus more strongly affected by β -casein polymorphism than by κ -casein polymorphism.

Protein content was most affected by κ -casein polymorphism; the composite genotypes A_1A_1BB , A_1A_1AB , and A_1A_2AB were associated with the highest protein content and those including the κ -casein E allele with the lowest (V).

Among those composite casein genotypes with the most favourable effect on milk and protein yields, milk yield was about 100 kg higher for genotype A_2A_2AB than for genotypes A_2A_2AA and A_1A_2AE . This indicated that the rare haplotype A_2B may have a strong favourable effect on milk and protein yields.

5.7.2 β - κ -casein haplotypes

The effect described above of β -casein polymorphism on milk and protein yields and fat content, and that of κ -casein polymorphism on protein content were apparent also when the overall effects of the casein haplotypes were estimated (VI). It is thus possible that the β -casein locus or genes linked to that locus affect milk and protein yields and fat content, and the κ -casein locus or genes linked to it affect protein content. The underlying reasons for the effects of casein polymorphism on milk-production traits are, however, unknown.

The haplotype A_2B , which had a favourable effect on milk and protein yields in VI, was further studied within the β - κ -casein genotype A_1A_2AB . This was done to evaluate whether the favourable association of haplotype A_2B with milk and protein yields was due to the effect of this allele combination or due to genes that are linked to the casein loci. Effects of the different haplotypes within composite genotype A_1A_2AE were also studied.

Within the genotype A_1A_2AB , the haplotype combination A_1A+A_2B produced 140 kg more milk ($P = 0.045$) with 0.03% units lower protein content ($P = 0.055$) than A_1B+A_2A . Within the β - κ -casein genotype A_1A_2AE , haplotype combination A_1A+A_2E was associated with 0.02% units higher protein content ($P = 0.098$) than the combination A_1E+A_2A . These results indicate that genes linked to the casein loci contribute to the variation in milk yield and protein content. Although an animal model was used to estimate the casein haplotype effects, the effects of these unknown genes could not be totally corrected for.

5.7.3 β -lactoglobulin genotypes

The β -lactoglobulin genotypes had a clear effect on first-lactation milk-production traits (V). The β -lactoglobulin A allele was associated with high milk and protein yields, and the B allele with high fat content.

6 DISCUSSION

The objective of this thesis was to study the possibilities of genetically improving the MCP of dairy cows. For this purpose, four separate data sets were collected, which were used for estimating 1) the genetic parameters for MCP, 2) milk protein polymorphism and its effect on MCP and milk-production traits, 3) occurrence of noncoagulating milk, 4) the association between coagulation properties of herd milk and cheese-making characteristics, and 5) differences in MCP between the main Finnish dairy breeds.

According to the results presented in I to VI, genetic improvement of MCP would be a well-supported means to increase the efficiency of cheese production. Improvement in MCP would be possible by selecting breeding animals directly for these traits or for properties associated with them.

6.1 Selection for MCP

Table 3: Estimates (est.) of heritability for milk coagulation time (R) and curd firmness (E_{30}) (underlined), and of genetic correlations between these traits and various milk-production characteristics.

	R	E_{30}	pH	DMY ¹	F-% ²	P-% ³	SCC ⁴	C-% ⁵	S ⁸
	est.	est.	est.	est.	est.	est.	est.	est.	
	(se) ⁶	(se)	(se)	(se)	(se)	(se)	(se)	(se)	
R	<u>0.22</u>	-0.96	0.40	0.02	-0.01	0.49	-0.06		Paper II
	(0.05)	(0.02)	(0.12)	(0.15)	(0.10)	(0.08)	(0.16)		
R	<u>0.27</u>		0.55		-0.91	-0.58	-0.03 ⁷		L ⁹
	(0.12)								
R	<u>0.27</u>		0.31	-0.23		0.13		-0.30	O ¹⁰
	(0.06)		(0.16)	(0.18)		(0.18)		(0.22)	
R	<u>0.38</u>		0.31	-0.26		-0.02		-0.50	O ¹¹
	(0.11)		(0.23)	(0.25)		(0.24)		(0.37)	
E_{30}		<u>0.40</u>	-0.30	-0.06	0.09	-0.24	0.18		Paper II
		(0.04)	(0.07)	(0.12)	(0.08)	(0.07)	(0.13)		
E_{30}		<u>0.30</u>	-0.33	0.03		0.13		0.48	O ¹⁰
		(0.07)	(0.20)	(0.18)		(0.17)		(0.18)	
E_{30}		<u>0.39</u>	-0.37	0.18		0.18		0.53	O ¹¹
		(0.12)	(0.28)	(0.26)		(0.23)		(0.31)	

¹Daily milk yield, kg, ²Fat content, %, ³Protein content.

⁴Somatic cell count, cells/ml. In II, SCC was logarithmically transformed.

⁵Casein content, %.

⁶Standard error of the estimate.

⁷Correlation between CT and the value for a viscometric reading.

⁸Source of the information.

⁹Lindström et al. (1984).

¹⁰Oloffs et al. (1992), 1874 Friesian cows, ¹¹Oloffs et al. (1992), 786 Angler cows.

Selection of breeding animals for their genetic ability to produce well-coagulating milk is possible; wide variation exists in MCP between cows, and a moderate proportion ($h^2 = 0.33$ to 0.62) of this variation is genetic (I, II). Heritability estimates in I and II agree rather well

with those presented by LINDSTRÖM et al. (1984), TERVALA et al. (1985), and OLOFFS et al. (1992) (Table 3).

The rather high repeatability estimates ($r = 0.57$ to 0.71) for MCP (I) indicate that even just a few measurements for these properties would give a good picture of the average coagulation ability of the milk of a cow. It would, in fact, be unnecessary to measure MCP more than three times during a lactation, because beyond that, the increase in the accuracy of the breeding value estimates for these properties would be negligible. The repeatability estimates presented in I were of the same magnitude as those presented by SCHAAR (1984), CAROLI et al. (1990) and TYRISEVÄ (1999a).

Noncoagulation of milk, which was observed as early as in the 1980s (TERVALA et al. 1983, 1985), was rather common in the FAy (I, II). Further, of a sample of about 4600 FAy cows collected in 1999, 13% produced noncoagulating milk (TYRISEVÄ et al. 1999b). Because such milk is poorly suited for cheese production (III), it is important to establish the factors causing it, and to seek means to decrease its occurrence.

6.1.1 Effect on other dairy traits

The genetic correlation between curd firmness and milk yield was low (Table 3), which was observed also in OLOFFS et al. (1992), and in a sample of 4600 FAy cows (TYRISEVÄ et al. 2000). As a consequence, direct selection for MCP may have no significant effect on milk yield, or on protein and fat yields, which strongly correlate with milk yield (TORNIAINEN 1991, ALBUQUERQUE et al. 1995).

Even though protein content and casein content have been shown to have a favourable phenotypic effect on milk coagulation ability (PAGNACCO and CAROLI 1987, VAN DEN BERG et al. 1992, Paper I), the estimates of the genetic associations between these characteristics have been inconsistent (Table 3). In II, high protein content of milk was unfavourably correlated with curd firmness, whereas no clear association existed between these traits in OLOFFS et al. (1992) or in TYRISEVÄ et al. (2000). In LINDSTRÖM et al. (1984), high protein content correlated favourably with milk coagulation time.

Even though protein content of milk, which correlates strongly with casein content of milk (HAYES et al. 1984), was not associated with MCP in OLOFFS et al. (1992), high casein content of milk correlated with favourable MCP in OLOFFS et al. (1992) (Table 3). In TYRISEVÄ et al. (2000), no strong correlation existed between MCP and casein content. It is thus unclear how breeding for well-coagulating milk would affect the protein content and protein composition.

No reliable estimate for the genetic correlation between curd firmness and somatic cell count of milk was obtained (Table 3), whereas in TYRISEVÄ et al. (2000), curd firmness correlated unfavourably with high somatic cell count. In LINDSTRÖM et al. (1984), milk coagulation time did not correlate with udder health. pH of milk, which correlates positively with somatic cell count, was negatively correlated with milk coagulation ability (Table 3), a fact observed also by LINDSTRÖM et al. (1984), OLOFFS et al. (1992), and TYRISEVÄ et al. (2000). Consequently, selection for MCP should have only a weakly favourable effect on udder health.

In this thesis, genetic associations between MCP and characteristics that describe the size, conformation, and fertility of dairy cows were not estimated. In addition, no estimates for these associations seem to exist in the literature.

6.1.2 Measurement for MCP

Genetic improvement of MCP by selection requires reliable measurement of MCP for the animals in a breeding population.

In Finland, 72% of the 380 000 dairy cows are included in milk recording, and 75% of these cows are FAY cows (Association of Rural Advisory Centres 1998). Consequently, the breeding population of the FAY consists of about 200 000 animals, and that of the FFR of about 70 000 animals.

The capacity of the current milk coagulation meters (Formagraph, Foss Electric A/S, and CRM48, Polo Trade, Monselice, Italy) to measure MCP is about 200 samples a day. Such instruments are thus unsuitable for routine measurement of MCP in milk recording, in which various dairy traits are measured several times during lactation. Further, production of the Formagraph instrument ended several years ago. The CRM48 instrument could, however, be used to measure MCP for a limited number of animals, e.g., bull dams and daughters of young AI-bulls.

Bull dams. About 800 cows are chosen as bull dam candidates every year. Measurement of MCP for these cows three times during lactation would thus mean several thousand measurements a year. Because wide variation exists in MCP between herds, and because of other systematic environmental factors (II, III), MCP should be measured also for herd mates of the bull dams.

Young AI-bulls. In Finland, about 220 FAY and about 70 FFR bull calves are chosen as AI-bull candidates every year. Because some 20% of these candidates are discarded for various reasons (e.g., growth and the quality of semen) before inseminations, about 200 of these bulls will be progeny tested.

For a well-inherited trait, records from about 50 daughters should be adequate for a reliable genetic evaluation of an AI-bull. To obtain breeding value estimates for MCP for young AI-bulls, milk samples of about 10 000 daughters and of their herd mates should thus be measured for MCP annually.

Consequently, to obtain breeding value estimates for MCP for bull dams or young AI-bulls, or both, a few CRM 48 instruments would be sufficient for all necessary measurements.

6.2 Selection for associated characteristics

6.2.1 Dairy traits

Based on the results in II, and in LINDSTRÖM et al. (1984), OLOFFS et al. (1992), and TYRISEVÄ et al. (2000), none of the routinely recorded dairy traits can be utilised to indirectly improve MCP.

Even though the genetic association between casein content and coagulation ability of milk in the FAY is weak (TYRISEVÄ et al. 2000), casein is important in cheese production (POLITIS and NG-KWAI-HANG 1988a and 1988b). Consequently, even though breeding for high casein content may have no strong effect on MCP, it may yet improve the usability of milk in cheese-making.

The unfavourable association between high pH and coagulation ability of milk indicates that breeding for low pH of milk may improve MCP. Both pH and casein content of milk are rather well-inherited traits (II), so genetic improvement of these properties may be possible. At the moment, routine measurement of these traits in milk recording is, however, impossible.

6.2.2 β -casein polymorphism

Even though the β -casein A₁ allele in the FAy and the rare β -casein B allele in the FFr were associated with somewhat better MCP than were the other alleles, the effect of β -casein polymorphism on these properties was small (Table 4). Selection based on β -casein polymorphism to improve MCP is thus unjustified. Further, selection for the β -casein A₁ allele would have an unfavourable effect on milk and protein yields. A favourable effect of the β -casein B allele on MCP was reported also by PAGNACCO and CAROLI (1987) and LODES et al. (1996a).

6.2.3 β -lactoglobulin polymorphism

β -lactoglobulin genotypes had no strong effect on MCP (Table 4), a fact observed also by TERVALA et al. (1983, 1985), PAGNACCO and CAROLI (1987), and VAN DEN BERG et al. (1992). In LODES et al. (1996a), the β -lactoglobulin B allele was, on the contrary, associated with the most favourable MCP, and in SCHAAR et al. (1985) with the most favourable cheese-making properties.

Selection for the β -lactoglobulin B allele may, however, have a favourable effect on cheese-making properties through its favourable effect on fat content (V, VI), casein content (I), and casein number (I). Because the β -lactoglobulin B allele is very common in the FAy (V), favouring this allele in selection would, however, be impractical in this breed.

The favourable effect of the β -lactoglobulin B allele on fat content has been observed also by NG-KWAI-HANG et al. (1984), ALEANDRI et al. (1990), BOVENHUIS et al. (1992), and ORTNER et al. (1995), and on casein content or casein number or both by MCLEAN et al. 1984, SCHAAR et al. (1985), NG-KWAI-HANG et al. (1986), LODES et al. (1997), and LUNDÉN et al. (1997).

6.2.4 κ -casein polymorphism

Effect on MCP. The κ -casein B allele had a favourable effect on MCP in I and II, in which an animal model was used to estimate the effect of milk protein polymorphism on these properties (Table 4). The favourable effect of the κ -casein B allele on MCP is thus apparent also after adjustment of the effect of this allele for the polygenic effects. Breeding for favourable MCP should thus be possible by favouring the κ -casein B allele. By current DNA-based techniques, genotyping of animals of both sexes and of all ages for milk proteins is possible. Milk protein genotypes could thus be easily determined for bull dam and AI-bull candidates among Finnish dairy cattle.

The favourable effect of the B allele has also been reported in several other studies, in which the least squares procedure was used (TERVALA et al. 1983, SCHAAR 1984, TERVALA et al. 1985, MARZIALI and NG-KWAI-HANG 1986, AALTONEN and ANTILA 1987, PAGNACCO and CAROLI 1987, DAVOLI et al. 1990, VAN DEN BERG et al. 1992, WALSH et al. 1995, and LODES et al. 1996a).

The κ -casein E allele had the most unfavourable effect on MCP (I, II), a finding observed also of LODES et al. (1996a). Another means of improving MCP is thus to select against the κ -casein E allele.

In the FFr, the κ -casein B allele was more frequent and the E allele rarer than in the FAy (Table 2). One possible reason for the difference in MCP between these breeds may thus be the difference in the frequency of the κ -casein alleles.

Table 4: Main associations of β -casein, κ -casein, and β -lactoglobulin (β -LG) alleles with coagulation and composition characteristics of milk and with milk-production traits. + indicates favourable effect on a trait, – unfavourable effect, and \downarrow indirect effect because of linkage disequilibrium between κ -casein and β -casein alleles. Any association within parentheses indicates a possible effect of the allele, and lack of a sign indicates no effect of the allele.

Trait	β -casein		κ -casein			β -LG		Paper
	A ₁	A ₂	A	B	E	A	B	
Curd firmness, mm				+	–			I
Curd firmness, mm	(+)	(–)	–	+	–			II
Curd firmness, mm	(+)	(–)	–	+	–			III
Milk yield, kg								II
Milk yield, kg	–	+		\downarrow	\downarrow	+	–	V, VI
Protein yield, kg	–	+		\downarrow	\downarrow	+	–	V, VI
Fat content, %								II
Fat content, %	+	–				–	+	V, VI
Protein content, %								II
Protein content, %				+	–			V, VI
Casein content, %						–	+	I
Casein number, %						–	+	I
κ -casein content, g/l				+				I

Because the κ -casein alleles were not associated with noncoagulation of milk (I, II), selection for these alleles would probably not alter the proportion of the cows producing milk with this defect. Based on the relationships observed between the individual cows that produce noncoagulating milk (II), it is, however, probable that other loci are involved in this phenomenon.

Effect on milk-production traits. That the κ -casein B allele alone has no significant effect on milk, protein, or fat yields (V, VI) was observed also by MCLEAN et al. (1984), LIN et al. (1986), ALEANDRI et al. (1990), BECH and KRISTIANSEN (1990), NG-KWAI-HANG et al. (1990), BOVENHUIS et al. (1992), TAHA and PUHAN (1993), RON et al. (1994), ORTNER et al. (1995), and SABOUR et al. (1996). According to LIN et al. (1989), VAN EENENNAAM and MEDRANO (1991), and MAO et al. (1992), the κ -casein B allele is associated with high milk yield, and, according to NG-KWAI-HANG et al. (1984), with high protein yield.

Because the κ -casein B allele was in linkage disequilibrium with the β -casein A₁ allele, and the latter had a clearly unfavourable effect on milk and protein yields (V to VI), selection for the κ -casein B allele to improve MCP would in most cases have an unfavourable effect on milk and protein yields. This fact would discourage selection for the B allele in the entire dairy population. Because the κ -casein E allele is also in linkage disequilibrium with the β -casein A₁ allele (V), selection against the E allele would instead have a favourable effect both on MCP, and on milk and protein yields and protein content of milk.

The unfavourable effect of the β -casein A₁ allele on milk and protein yields and the favourable effect of this allele on fat content (V to VI) has been reported in some of the other studies (NG-KWAI-HANG et al. 1984, LIN et al. 1986, BECH and KRISTIANSEN 1990, NG-KWAI-HANG et al. 1990, ORTNER et al. 1995, VELMALA et al. 1995), but not in ALEANDRI et al. (1990), and LIN et al. (1989).

Findings concerning the effect of κ -casein polymorphism on protein content of milk were inconsistent; the κ -casein B allele was associated with high protein content, and the E allele with low protein content in studies with large data (V to VI), but not with less data (I, II). Favourable effect of the κ -casein B allele on protein content has been observed also in VAN EENENNAAM and MEDRANO (1991), BOVENHUIS et al. (1992), MAO et al. (1992), and BOVENHUIS and WELLER (1994), but not in SCHAAR et al. (1985) or LODES et al. (1997).

The favourable effect of the κ -casein B allele on κ -casein content (Table 4), which partly explained the favourable effect of this allele on MCP (I), has been reported also by MCLEAN et al. (1984), KROEKER et al. (1985), NG-KWAI-HANG et al. (1987), VAN DEN BERG et al. (1992), LODES et al. (1996b), and BOBE et al. (1999). Other possible causes for the differences in MCP between the κ -casein alleles are casein micelle size, citrate content, calcium content, and electric charge (SCHAAR 1984, PAGNACCO and CAROLI 1987, POLITIS and NG-KWAI-HANG 1988c, VAN DEN BERG et al. 1992, and LODES et al. 1996b).

Effect on other dairy traits. In addition to high protein and fat production, the current breeding objectives of the Finnish dairy cattle are good fertility and udder health. Milk protein genotypes had no significant effect on such heifer and first-lactation reproduction traits of FAY cows (RUOTTINEN et al. 1998). Effects of casein genotypes on udder health of FAY and FFr cows have not yet been estimated. According to ATROSHI et al. (1982), β -lactoglobulin polymorphism in the FAY had no effect on milk somatic cell count.

Frequencies of the κ -casein B and E alleles. In addition to its linkage disequilibrium with the β -casein A₁ allele, the rarity of the κ -casein B allele in Finnish dairy cattle would restrict selection for it. For example, based on the frequencies of the κ -casein alleles in the FAY (V), 30 bulls (14%) of 220 FAY candidate AI-bulls would be heterozygous for the κ -casein B allele, and only 2 bulls (1%) homozygous for it. Among 500 FAY bull dam candidates, the corresponding numbers would be 70 and 5. Pre-selection of AI-bull or bull dam candidates based on the κ -casein B allele would thus be difficult.

On the other hand, if one were to seek 220 FAY AI-bull candidates and 500 FAY bull dam candidates that carried at least one copy of the κ -casein B allele, about 1600 bull calves and 3500 bull dam candidates should be selected for milk protein genotyping. This would result in a decrease in the quality standards and in the selection intensity of the breeding animals for the other traits included in the breeding goal.

Because the κ -casein E allele is rather common in the FAY (IV), the potential exists for genetic improvement of MCP in this breed by selection against the E allele. In the FFr, the κ -casein E allele is rather rare, so selection against it would have a small effect on these traits.

7 CONCLUSIONS AND IMPLICATIONS

According to the findings presented in I to VI, genetic improvement of MCP is a well-justified means to increase the efficiency of cheese production. Improvement of MCP is possible by selection of breeding animals for these traits or for properties associated with them (Figure 4).

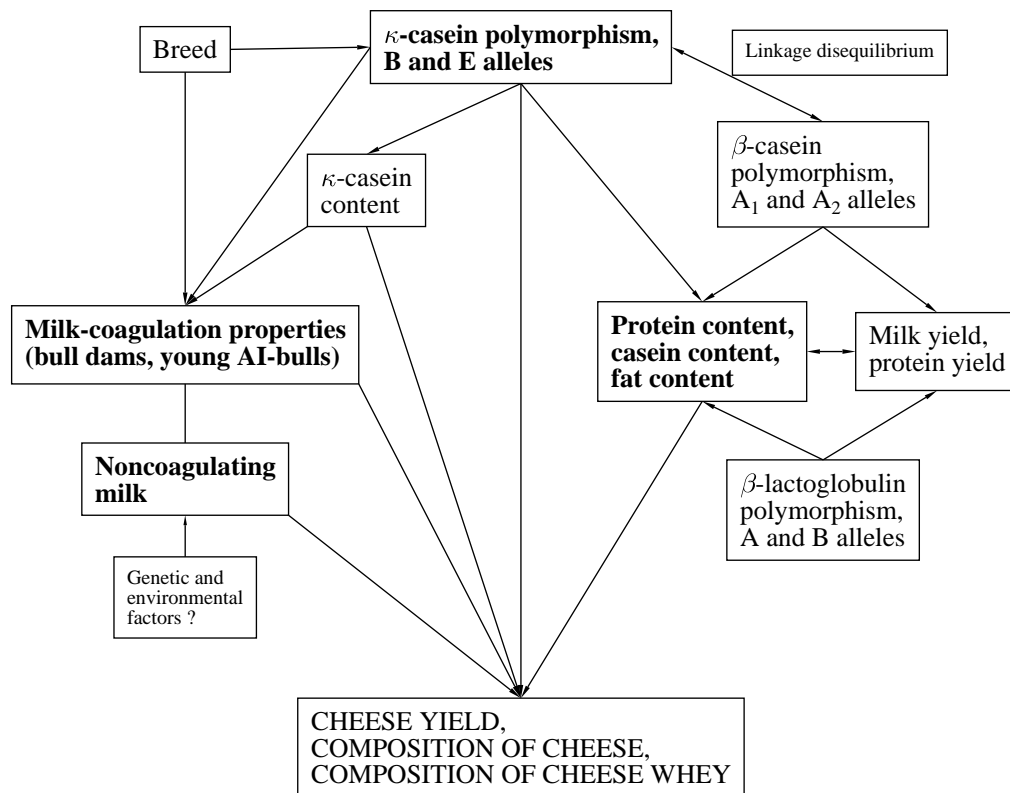


Figure 4: Diagram of established interrelationships between milk protein polymorphisms, MCP, milk-production traits, composition of milk, and cheese-making properties (I to VI). Characteristics useful in genetic improvement of milk-coagulation or cheese-making characteristics or both in boldface.

Because measurement of the three MCP, R, K₂₀ and E₃₀ (Figure 1), is labourious and slow, the current possibilities of directly breeding for these properties in the entire dairy population are limited. Because genetic improvement of MCP is most effective by direct selection for these properties, breeding values for MCP could, however, be estimated for an important group of animals in the dairy population: bull dams and daughters of young AI-bulls (Figure 4).

Because none of the routinely recorded dairy traits strongly correlate with MCP, breeding for these traits does not markedly affect MCP. Because of their important role in the cheese-

making process, breeding for high casein content and fat content could, however, have a favourable effect.

The κ -casein alleles have a clear effect on coagulation properties, protein composition, and cheese-making properties of milk (Figure 4). Because of the rarity of the favourable B allele, and linkage disequilibrium between the κ -casein B allele and β -casein A₁ allele, selection for the B allele in the entire Finnish dairy cattle would be complicated.

The κ -casein E allele, which is rather common in the FAy, is associated with poor MCP and low protein content, and, because of the linkage disequilibrium between the E allele and β -casein A₁ allele, also with low milk and protein yields. Selection against this allele would thus have a favourable effect both on MCP and on important milk-production traits. Because it is possible that genes linked to the casein loci affect milk yield and protein content of milk (VI), it would, however, be important to try to locate these genes and to estimate their influence on selection based on κ -casein polymorphism.

Noncoagulating milk, which was not associated with milk protein polymorphism or composition of milk, had an unfavourable effect on cheese-making properties. The environmental and genetic factors causing noncoagulation of milk should be established.

In conclusion, at present, information on breeding values for MCP and on κ -casein polymorphism for young AI-bulls and bull dams could be useful in genetically improving MCP in Finnish dairy cattle. Although such information would not serve as an official selection criterion of these animals, it could be made available to farmers who provide milk for cheese production, either at their own farms or at a commercial cheese plant.

7.1 Suggestions for future study

The findings presented in I to VI give an evaluation of the current possibility of genetically improving MCP. Additional information about MCP, and about the factors affecting them is, however, necessary to gain a more complete picture of these properties and of how to breed for them.

- The capacity of the current milk-coagulation meters is insufficient for measuring MCP for all cows in milk recording. Development of equipment for this purpose may thus be worth considering.
- The association between coagulation and cheese-making properties of milk should be studied based on milks from several herds (e.g., those providing milk for a cheese plant). It would thus be possible to estimate in more detail the association between the MCP and efficiency of cheese-making, and the economic advantage of genetic improvement of MCP.
- To decrease the occurrence of noncoagulating milk in the FAy, the hypothetical genetic factors causing it must be discovered.
- To fully understand the effect of milk protein polymorphism on MCP and milk-production traits, the association between genetic protein polymorphism and expression of the milk protein genes need further study.
- Reasons for differences in MCP between the two main Finnish dairy breeds should be established.

- Effects of milk protein genotypes on size, conformation, health, and management traits should be studied to gain a complete picture of the effect of milk protein polymorphism on dairy traits.
- Although the effect of utilising information on κ -casein polymorphism in selection of breeding animals for dairy production has been estimated in a few studies (GIBSON et al. 1990, PEDERSEN 1991, LIN et al. 1992, BOVENHUIS and DE BOER 1994, and PABST 1997), advantages and disadvantages of using this information and also information on MCP in the selection of Finnish dairy animals need to be estimated.

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