Genetic Parameters for Major Milk Fatty Acids and Milk Production Traits of Dutch Holstein-Friesians

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ABSTRACT

The objective of this study was to estimate genetic parameters for major milk fatty acids and milk production traits. One morning milk sample was collected from 1,918 Holstein-Friesian heifers located in 398 commercial herds in the Netherlands. Each sample was analyzed for total percentages of fat and protein, and for detailed fatty acid percentages (computed as fatty acid weight as a proportion of total fat weight). Intraherd heritabilities were high for C4:0 to C16:0, ranging from 0.42 for C4:0 to 0.71 for C10:0. Saturated and unsaturated C18 fatty acids had intraherd heritability estimates of approximately 0.25, except for C18:2 *cis*-9, *trans*-11, which was 0.42. Standard errors of the heritabilities were between 0.07 and 0.12. Genetic correlations were high and positive among C4:0 to C14:0, as well as among unsaturated C18, but correlations of C4:0 to C14:0 with unsaturated C18 were generally weak. The genetic correlation of C16:0 with fat percentage was positive (0.65), implying that selection for fat percentage should result in a correlated increase of C16:0, whereas unsaturated C18 fatty acids decreased with increasing fat percentage (−0.74). Milk fat composition can be changed by means of selective breeding, which offers opportunities to meet consumer demands regarding health and technological aspects.

Key words: fatty acid, milk composition, genetic parameter, dairy cattle

INTRODUCTION

Milk fat contains many nutrients necessary for humans, including fat-soluble vitamins, energy, and bioactive lipids (German and Dillard, 2006). During the last decennia, however, milk fat consumption has become negatively associated with human health (Bitman et al., 1995; Jensen, 2002). Cow milk fat contains a

relatively low proportion of unsaturated fatty acids (UFA) and a relative high proportion of LDL-cholesterol-increasing fatty acids (FA), mainly C14:0 and C16:0 (German and Dillard, 2006). Both have been associated with increased levels of cholesterol and increased risk of cardiovascular disease (Maijala, 2000; Jensen, 2002). In the past, some studies have questioned whether changing milk fat composition could result in a significant improvement for human health (Gibson, 1991; Maijala, 1995). In recent years, however, market attention has increasingly focused on improving the health aspects of (dairy) products. Possibilities of changing milk fat composition by genetically altering proportions of FA are therefore of interest. To study these possibilities, genetic parameters for FA have to be estimated.

Soyeurt et al. (2006a), in studying 600 milk samples of 275 animals from 5 breeds, found differences in FA proportions among breeds of cattle. Lawless et al. (1999) studied approximately 25 animals per breed and found a relatively high amount of C16:0 and a slightly lower amount of C18:0 in Holstein-Friesians, compared with Normande and Montbeliarde breeds.

Renner and Kosmack (1974b) studied 243 cows originating from 10 sires and found evidence for the existence of within-breed genetic variation in milk fat composition. Karijord et al. (1982) and Famula et al. (1995) also estimated within-breed genetic variation for milk fat composition. The study by Karijord et al. (1982) consisted of approximately 3,000 animals and 7,000 samples, and Famula et al. (1995) studied 523 animals and one sample per animal. Information on within-breed genetic variation in milk fat composition, however, is still limited. The aim of this study was to estimate genetic parameters for major milk FA and to study the relationship of major milk FA with milk production traits in the Dutch Holstein-Friesian population.

MATERIALS AND METHODS

Animals and Milk Samples

This study was part of the Milk Genomics Initiative, which focuses on genetic aspects of milk composition.

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The Milk Genomics Initiative was designed to have approximately 2,000 cows descending from a number of selected bulls; 50 young bulls were aimed to have 20 daughters each, and 5 proven bulls were aimed to have 200 daughters each. The pedigree was the leading reason for the choice of farms; all the farms in the database of the NRS (Arnhem, the Netherlands) with at least one proven bull heifer and one test bull heifer were invited to participate in the study. When 2,000 animals were assigned to the study, no further farms were selected.

To study milk fat composition, data were available from 1,918 cows on 398 commercial herds in the Netherlands. Of those cows, 843 cows were sired by the 50 young bulls, 888 cows by the 5 proven bulls, and 187 cows by 46 other proven bulls, to have at least 3 cows per farm. Each cow was more than 87.5% Holstein-Friesian, and was between 63 and 263 DIM of the first lactation.

One milk sample of 500 mL per cow was collected between February and March 2005. Cows were milked twice daily, but only the morning milk was collected for the study to ensure the quality of the samples. Milk was cooled to 4°C within 3 h after sampling and transported to the laboratory the same morning. Sample bottles contained sodium azide (0.03% wt/wt) for conservation.

Analysis of Milk Samples

Milk fat (butter) was extracted from approximately 400 mL of milk, keeping the remaining 100 mL for other analyses. Fatty acid methyl esters were prepared from milk fat as described in ISO Standard 15884 (International Organization for Standardization-International Dairy Federation, 2002b). The methyl esters were analyzed by gas chromatography according to the 100% FA methyl ester method (International Organization for Standardization-International Dairy Federation, 2002a) with a 100-m polar column (Varian Fame Select CP 7420, Varian Inc., Palo Alto, CA) at the laboratory of the Netherlands Controlling Authority for Milk and Milk Products (Leusden, the Netherlands). The FA were identified and quantified by comparing the methyl ester chromatograms of the milk fat samples with the chromatograms of pure FA methyl ester standards, and were measured as the weight proportion of total fat weight. The chromatograms resulted in approximately 130 measurable FA peaks, of which approximately one-third could be identified. Of these, 16 major FA were used for the estimation of genetic parameters in the present study: the even-numbered FA C4:0 to C18:0; 5 identified C18:1 isomers; C18:2 *cis*-9,12; C18:3 *cis*-9,12,15; and conjugated linoleic acid (CLA) *cis*-9, *trans*-

Table 1. Trait definition: Groups of fatty acids

Group	Content
C6-12	C6:0, C8:0, C10:0, C12:0
C14-16	C14:0, C16:0
C18u	C18:1 <i>trans</i> -4–8, C18:1 <i>trans</i> -9, C18:1 <i>trans</i> -11, C18:1 <i>cis</i> -9, C18:1 <i>cis</i> -11, C18:2 <i>cis</i> -9,12, C18:3 <i>cis</i> -9,12,15
CLA	Conjugated C18:2 <i>cis</i> -9, <i>trans</i> -11
Ratio	Ratio between saturated and unsaturated fatty acids
Saturated	C4:0, C5:0, C6:0, C7:0, C8:0, C9:0, C10:0, C11:0, C12:0, C13:0, C14:0, C15:0, C16:0, C17:0, C18:0
Unsaturated	C10:1, C12:1, C14:1, C16:1, C18u, CLA

11. These 16 FA comprised 89% of the total fat. In addition to the individual FA, a number of FA groups were defined based on their potential effect on human health (German and Dillard, 2006; Table 1): a “neutral” group (**C6-12**) containing C6:0, C8:0, C10:0, and C12:0; a “negative” group (**C14-16**) containing C14:0 and C16:0; and a “positive” group (**C18u**) containing all unsaturated C18 that were part of the data set. Data were analyzed as weight proportion. To calculate the ratio of SFA:UFA, 11 additional monounsaturated FA and odd-chain FA were included.

Percentages of fat and protein were determined from a 10-mL milk subsample by infrared spectroscopy by using a Fourier-transformed interferogram (MilkoScan FT 6000, Foss Electric, Hillerød, Denmark) at the certified laboratory of the Milk Control Station (Zutphen, the Netherlands).

In total, 1,918 records were analyzed for fat and protein percentages and fat composition. The NRS supplied the corresponding morning test-day milk yield of the samples; 1,783 NRS records matched our samples. Data for milk yield, fat yield, and protein yield was missing for 135 cows.

Statistical Analyses

Variance components and genetic parameters were estimated by using an Animal model in ASReml (Gilmour et al., 2002):

$$y_{ijklmn} = \mu + b_1 \times \text{dim}_i + b_2 \times e^{-0.05 \times \text{dim}} + b_3 \times \text{afc}_j + b_4 \times \text{afc}_j^2 + \text{season}_k + \text{scode}_l + \text{herd}_m + A_n + e_{ijklmn}, \quad [1]$$

where y_{ijklmn} is the dependent variable corresponding to the observation of animal n in herd m , with scode_l age at first calving j in season k , and DIM i ; μ is the general mean; dim_i is the covariate for DIM, modeled with a Wilmink curve (Wilmink, 1987); afc_j is the covariate describing the effect of age at first calving; season_k

is the fixed effect with 3 classes for season of calving, summer (June to August 2004), autumn (September to November 2004), and winter (December 2004 to February 2005); $scode_1$ is the fixed effect accounting for possible differences between the groups of proven bull daughters and young bull daughters; $herd_m$ is the random effect defining groups of animals sampled in the same herd; A_n is the random additive genetic effect of animal n ; and e_{ijklmn} is the random residual effect.

Relationships between individuals were accounted for, and the total pedigree consisted of 26,300 animals and was supplied by NRS (Arnhem, the Netherlands). Heritabilities were estimated by using univariate analyses. We defined two heritabilities:

$$h_{IH}^2 = \frac{\sigma_A^2}{\sigma_A^2 + \sigma_e^2} \quad [2]$$

where σ_A^2 is the additive genetic variation, and σ_e^2 is the residual variation. Heringstad et al. (2006) referred to this heritability as the intraherd heritability. The intraherd heritability is the parameter that is required to predict selection responses of alternative breeding strategies. The ratio of the additive genetic over the total phenotypic variance was defined as

$$h^2 = \frac{\sigma_A^2}{\sigma_A^2 + \sigma_{herd}^2 + \sigma_e^2} \quad [3]$$

where σ_{herd}^2 is the herd test-day variation.

The proportion of variance attributable to herd reflects the relative importance of herd effects such as feed, hygiene, and management, and could be estimated as

$$h_{herd} = \frac{\sigma_{herd}^2}{\sigma_A^2 + \sigma_{herd}^2 + \sigma_e^2} \quad [4]$$

To compare the relative importance of genetic and herd effects, the ratio $\sigma_A^2/\sigma_{herd}^2$ was calculated.

Genetic correlations were estimated by using bivariate analyses and model [1] as

$$r_g = \frac{\sigma_{A1,A2}^2}{\sqrt{(\sigma_{A1}^2 \times \sigma_{A2}^2)}}, \quad [5]$$

where $\sigma_{A1,A2}^2$ is the additive genetic covariance between trait 1 and trait 2, and σ_{A1}^2 and σ_{A2}^2 are the additive genetic variance of traits 1 and 2.

A principal component analysis PROC VARCLUS, in combination with PROC TREE in SAS (SAS 9.1, SAS

Institute, 1999) was used to graphically visualize genetic correlations among major FA.

RESULTS

Mean and Coefficient of Variation

Mean, coefficient of variation, and 5 and 95% quartiles for individual FA, groups of FA, and milk production traits are shown in Table 2. The results showed considerable variation in milk fat composition among cows. Short-chain FA (C4:0 to C12:0) averaged 15% of the total fat, medium-chain FA (C14:0 and C16:0) averaged 44%, and C18:0 averaged 8%. The FA C18:1 *cis*-9, the most prominent among the UFA, averaged 18%, and the other UFA averaged 3.5%. The total FA in Table 2 made up approximately 89% of the total fat; the remaining 11% consisted of a large number of FA present in small amounts. These FA included unsaturated medium-chain FA, odd-chain FA, branched-chain FA, and several long-chain FA.

The ratio of saturated FA (SFA) to UFA averaged 2.8, indicating that approximately 74% of the fat was saturated. This number slightly overestimates the true ratio of SFA:UFA, because the ratio was based on only 27 major FA, whereas trace amounts of some FA, mainly UFA, were not taken into account.

The coefficients of variation for individual FA ranged from 7% for C6:0 to 28% for C18:2. A low coefficient of variation (approximately 10%) was found for most saturated FA (C4:0 to C18:0), and a higher coefficient of variation (approximately 25%) was found for most unsaturated C18 FA. The highest coefficient of variation was found for long-chain polyunsaturated FA: 28% for C18:2, and 27% for C18:3. The coefficient of variation for the entire C18u group was only 11%.

Heritability and Proportion of Variance Attributable to Herd

Intraherd heritabilities for individual FA, groups of FA, and milk production traits are shown in Table 3. High intraherd heritabilities of 0.42 to 0.71 were found for C4:0 to C16:0. Intraherd heritabilities of 0.22 to 0.35 were found for both saturated and unsaturated C18 FA, but for CLA *cis*-9, *trans*-11, heritability was 0.42. Intraherd heritabilities for the groups C6-12 (0.67) and C18u (0.26) were in line with the results for individual FA. For C14-16, intraherd heritability was rather low (0.16) compared with intraherd heritabilities of 0.59 for C14:0 and 0.43 for C16:0. Standard errors of intraherd heritability estimates were between 0.07 and 0.12.

Results for the proportion of variance explained by herd are also shown in Table 3. Differences among herds most likely represent differences in feeding regi-

Table 2. Number (n), mean,¹ SD, CV, and 5 and 95% quartile for fatty acids, groups of fatty acids,² and milk production traits, measured on a test-day morning milk sample of 1,918 cows in first lactation

Trait	n	Mean	SD	CV, %	5% quartile	95% quartile
Individual fatty acids						
C4:0	1,918	3.50	0.27	8	3.05	3.96
C6:0	1,918	2.23	0.17	7	1.95	2.48
C8:0	1,918	1.37	0.14	10	1.14	1.58
C10:0	1,918	3.03	0.43	14	2.34	3.73
C12:0	1,918	4.11	0.69	17	3.02	5.35
C14:0	1,918	11.62	0.91	8	10.03	13.01
C16:0	1,918	32.61	2.81	9	28.07	37.11
C18:0	1,918	8.73	1.42	16	6.55	11.07
C18:1 <i>cis</i> -9	1,918	18.02	2.09	12	14.92	21.76
C18:1 <i>cis</i> -11	1,918	0.41	0.11	27	0.26	0.61
C18:1 <i>trans</i> -4–8	1,918	0.21	0.05	24	0.15	0.29
C18:1 <i>trans</i> -9	1,918	0.15	0.03	20	0.11	0.20
C18:1 <i>trans</i> -11	1,918	0.77	0.20	26	0.49	1.13
C18:2 <i>cis</i> -9,12	1,918	1.20	0.29	24	0.81	1.68
C18:2 <i>cis</i> -9, <i>trans</i> -11 (CLA ³)	1,918	0.39	0.11	28	0.25	0.59
C18:3 <i>cis</i> -9,12,15	1,918	0.41	0.11	27	0.27	0.61
Groups of fatty acids						
C6-12	1,918	10.74	1.23	11	8.70	12.72
C14-16	1,918	44.24	2.78	6	39.51	48.47
C18u	1,918	21.58	2.43	11	18.19	26.16
Ratio ⁴ SFA:UFA	1,918	2.80	0.37	13	2.17	3.38
Milk production traits						
Fat, %	1,918	4.36	0.70	16	3.33	5.48
Protein, %	1,918	3.51	0.30	9	3.04	4.01
Fat yield, kg	1,783	0.58	0.11	19	0.40	0.76
Protein yield, kg	1,783	0.47	0.09	19	0.33	0.61
Milk yield, kg	1,783	13.5	2.7	20	9.0	18.1

¹For each fatty acid, mean = mean fatty acid as a proportion (wt/wt) of the total fat fraction of 100%.

²Groups are defined in Table 1.

³CLA = conjugated linoleic acid.

⁴SFA = saturated fatty acids; UFA = unsaturated fatty acids.

mens among herds, but the effects of other management factors also cannot be excluded. Variance attributable to herd was lower for saturated FA (C4:0 to C18:0, approximately 0.20), than for unsaturated C18 FA (approximately 0.50). This difference, however, was not found when comparing different groups of FA: for C6-12, herd explained 27% of the variation, whereas for C18u, herd explained 31%.

The ratio of genetic variance to variance attributable to herd ($\sigma_A^2/\sigma_{herd}^2$; Table 3) gives the relative importance of genetic vs. herd effects. For C4:0 to C18:0, genetic effects were generally larger than herd effects, whereas for unsaturated C18, herd effects were larger than genetic effects.

Genetic Correlations Among Individual FA

Genetic correlations among individual FA are shown in Table 4. The C4:0 had a moderate negative correlation with most other FA. The C6:0 to C14:0 FA were positively correlated (0.34 to 0.96), with a weak correlation of 0.08 for C6:0 with C14:0. The C16:0 showed negative correlations with all studied FA except for

C4:0, C6:0, and C18:0. The C18:0 also showed negative correlations with most other FA, but the correlations were weak. The unsaturated C18 FA were positively correlated (0.25 to 0.99), with a weak correlation of 0.12 for C18:1 *cis*-9 with C18:1 *trans*-11. A clustering tree to visualize the genetic correlations among FA is shown in Figure 1. Eight clusters explained more than 90% of the variance. The figure shows the clustering of C6:0 to C14:0 in one group and the clustering of unsaturated C18 in another group. This reflects the high correlations within these groups. The C4:0 and C16:0 did not cluster in one of these 2 main groups.

Genetic Correlations Among Groups of FA

Genetic correlations among the groups of FA [C4:0, C6-12, C18:0, C18u, CLA *cis*-9, *trans*-11, and the ratio of SFA:UFA] are shown in Table 5. The C4:0 shows moderate correlations with other FA, as does C6-12. Both C14-16 and C18:0 showed negative correlations with (unsaturated) C18 and CLA *cis*-9, *trans*-11, and a positive correlation with the ratio of SFA:UFA. The positive correlation was expected, because C14-16 and

Table 3. Genetic, residual, and herd variation, heritabilities,¹ proportion of variance explained by herd,² and ratio of genetic over herd variance for individual fatty acids, groups of fatty acids,³ and milk production traits, measured on a test-day morning milk sample of 1,918 cows in first lactation

Trait	σ_A^2	σ_E^2	σ_{herd}^2	h_{IH}^2	h^2	h_{herd}	$\sigma_A^2/\sigma_{herd}^2$
Individual fatty acids							
C4:0	0.027	0.038	0.013	0.42	0.35	0.17	2.09
C6:0	0.011	0.012	0.004	0.46	0.39	0.16	2.39
C8:0	0.010	0.006	0.004	0.61	0.48	0.20	2.42
C10:0	0.107	0.044	0.047	0.71	0.54	0.24	2.30
C12:0	0.177	0.106	0.217	0.63	0.35	0.43	0.81
C14:0	0.433	0.296	0.157	0.59	0.49	0.18	2.75
C16:0	2.458	3.296	2.298	0.43	0.31	0.29	1.07
C18:0	0.374	1.218	0.382	0.23	0.19	0.19	0.98
C18:1 <i>cis</i> -9	0.770	2.340	1.204	0.25	0.18	0.28	0.64
C18:1 <i>cis</i> -11	0.001	0.005	0.005	0.22	0.12	0.43	0.29
C18:1 <i>trans</i> -4–8	0.0004	0.0008	0.0013	0.35	0.18	0.49	0.35
C18:1 <i>trans</i> -9	0.0001	0.0004	0.0005	0.22	0.11	0.50	0.21
C18:1 <i>trans</i> -11	0.005	0.013	0.023	0.28	0.12	0.55	0.22
C18:2 <i>cis</i> -9,12	0.010	0.029	0.040	0.26	0.13	0.51	0.25
C18:2 <i>cis</i> -9, <i>trans</i> -11 (CLA ⁴)	0.003	0.004	0.006	0.42	0.21	0.49	0.43
C18:3 <i>cis</i> -9,12,15	0.001	0.003	0.007	0.26	0.09	0.64	0.14
Groups of fatty acids							
C6-12	0.785	0.385	0.437	0.67	0.49	0.27	1.81
C14-16	0.814	4.142	2.624	0.16	0.11	0.35	0.34
C18u	1.045	2.983	1.770	0.26	0.18	0.31	0.59
Ratio ⁵ SFA:UFA	0.027	0.071	0.038	0.28	0.20	0.28	0.71
Milk production trait							
Fat, %	0.239	0.231	0.038	0.51	0.47	0.07	6.25
Protein, %	0.046	0.025	0.017	0.65	0.53	0.19	2.76
Fat yield, kg	0.004	0.006	0.003	0.39	0.29	0.24	1.21
Protein yield, kg	0.001	0.004	0.003	0.23	0.14	0.37	0.39
Milk yield, kg	2.036	2.951	1.930	0.41	0.29	0.28	1.06

¹ $h_{IH}^2 = \sigma_A^2/(\sigma_A^2 + \sigma_e^2)$, SE between 0.07 and 0.12. $h^2 = \sigma_A^2/(\sigma_A^2 + \sigma_{herd}^2 + \sigma_e^2)$, SE between 0.07 and 0.12.

² $h_{herd} = \sigma_{herd}^2/(\sigma_A^2 + \sigma_{herd}^2 + \sigma_e^2)$, SE approximately 0.03.

³Groups are defined in Table 1.

⁴CLA = conjugated linoleic acid.

⁵SFA = saturated fatty acids; UFA = unsaturated fatty acids.

C18:0 are the main saturated fractions, so a relative increase will also increase the ratio. The C18u and CLA *cis*-9, *trans*-11 were highly correlated (0.71), and both showed a strong negative correlation with the ratio of SFA:UFA, because they are the main unsaturated fractions.

Genetic Correlations of FA with Production Traits

Genetic correlations of individual FA and groups of FA with milk production traits are shown in Table 6. Fat percentage showed a positive correlation with C16:0 (0.65), and negative correlations with C14:0 (–0.43) and all unsaturated C18 FA (–0.43 to –0.78). Results were similar for the correlations of fat percentage with groups of FA: fat percentage showed a positive correlation with C14-16 (0.65) and a strong negative correlation with C18u (–0.74).

Protein percentage and fat yield also had positive correlations with C16:0 and C14-16, and negative correlations with all unsaturated C18 FA. For protein yield

and milk yield, however, results were opposite: a negative correlation with C14-16 and moderate to strong positive correlations with all unsaturated C18 FA.

DISCUSSION

The results of the current study show that there is considerable genetic variation for proportions of FA, with genetic variation being high for C4:0 to C16:0 and moderate for C18 FA. The moderate coefficient of variation in combination with a moderate to high intraherd heritability indicates that FA proportions can be changed by genetic selection. The genetic correlations suggest that the individual FA react differently to selection. Selection on fat percentage seems to have little effect on C6-12, whereas it is expected to lead to an increase in C14-16 and a decrease in C18u.

Samples

In the current study, one morning milk sample per cow was analyzed. Only morning milk was used to en-

Table 4. Genetic correlations¹ between individual fatty acids, measured on a test-day morning milk sample of 1,918 cows in first lactation

Fatty acid	C4:0	C6:0	C8:0	C10:0	C12:0	C14:0	C16:0	C18:0	C18:1 <i>cis</i> -9	C18:1 <i>cis</i> -11	C18:1 <i>trans</i> -4-8	C18:1 <i>trans</i> -9	C18:1 <i>trans</i> -11	C18:2 <i>cis</i> -9,12	C18:3 <i>cis</i> -9,12,15	CLA ²
C6:0	0.48															
C8:0	-0.05	0.82														
C10:0	-0.40	0.55	0.92													
C12:0	-0.56	0.34	0.81	0.96												
C14:0	-0.49	0.08	0.50	0.72	0.76											
C16:0	0.52	0.15	-0.33	-0.26	-0.29	-0.84										
C18:0	0.15	-0.01	-0.23	-0.35	-0.35	-0.34	0.27									
C18:1 <i>cis</i> -9	-0.11	-0.58	-0.31	0.39	0.51	0.13	-0.37	-0.41								
C18:1 <i>cis</i> -11	-0.62	-0.31	0.11	0.39	0.39	0.02	-0.79	0.02	0.35							
C18:1 <i>trans</i> -4-8	-0.32	-0.38	-0.21	0.01	0.14	0.42	-0.51	0.05	0.25	0.47						
C18:1 <i>trans</i> -9	-0.49	-0.36	-0.01	0.20	0.26	0.46	-0.74	-0.44	0.63	0.59	0.66					
C18:1 <i>trans</i> -11	-0.12	0.05	0.07	0.09	0.13	0.12	-0.38	0.05	0.12	0.40	0.69	0.78				
C18:2 <i>cis</i> -9,12	-0.23	0.06	0.33	0.42	0.45	0.61	-0.79	-0.27	0.41	0.33	0.52	0.86	0.63			
C18:3 <i>cis</i> -9,12,15	-0.31	-0.28	-0.12	0.02	0.09	0.31	-0.61	-0.36	0.62	0.55	0.73	0.99	0.94	0.90		
C18:2 <i>cis</i> -9, <i>trans</i> -11 (CLA) ²	-0.14	-0.10	0.02	0.08	0.14	0.33	-0.59	-0.58	0.54	0.35	0.55	0.87	0.76	0.68	0.95	

¹SE between 0.01 and 0.26. Most SE were between 0.12 and 0.21, with strong correlations having SE between 0.01 and 0.10.²CLA = conjugated linoleic acid.

sure a quick transport from the farm to the laboratory. Usually, however, milk production data are analyzed by using a mixed evening-morning sample. This difference could have affected the obtained results. A study by Morris et al. (2005), using an interval of 8 h:16 h, showed high correlations of approximately 0.6 to 0.9 for proportions of FA between the evening and morning samples.

Chromatograms of the milk fat samples showed a total of 133 different peaks. About one-third of these peaks could be identified. The current study focused on the identified major FA, constituting approximately 90% of total fat, which were even-chain FA (C4:0 to C18:0); 5 monounsaturated C18:1; C18:2 *cis*-9,12; C18:3 *cis*-9,12,15; and the CLA isomer *cis*-9, *trans*-11. Because FA were measured as weight proportion, SFA and UFA were dependent variables and were analyzed as the ratio of SFA:UFA.

Heritabilities

In the current study, the effect of herd was modeled as a random effect, because our interest was also in variation attributable to herd effects. Most studies model herd as a fixed effect. Modeling herd as fixed or random effect has, among others, consequences for the total phenotypic variance, and consequently for the heritability estimates. We also performed an analysis in which we took herd as a fixed effect. The heritabilities that were obtained from these analyses (results not shown) were very similar to what was termed the intraherd heritability (h^2_{IH}). This makes the intraherd heritability reported in the current study comparable to the heritability reported by other studies that modeled herd as a fixed effect.

Renner and Kosmack (1974b) and Karijord et al. (1982) estimated heritabilities for short-chain FA between 0.13 and 0.26, for medium-chain FA between 0.06 and 0.11, and for unsaturated C18 at 0.04 (Renner and Kosmack, 1974b), which were lower than estimates found in the current study. Both studies (Renner and Kosmack, 1974b; Karijord et al., 1982) also found low heritabilities for milk yield (0.36 and 0.09) and fat percentage (0.28 and 0.09) compared with the current study. The milk production trait heritabilities found in this study were, however, in line with more recent literature (e.g., Vos and Groen, 1998; Ikonen et al., 1999; Ojala et al., 2004).

Short-Chain vs. Long-Chain FA

Intraherd heritability for C4:0 to C14:0 (approximately 0.60) was higher than for unsaturated C18 (approximately 0.25), which is in agreement with the find-

Figure 1. Cluster tree based on principal component analysis of genetic correlations among individual fatty acids. Eight clusters explained more than 90% of the variation (dotted line).

ings of Renner and Kosmack (1974b) and Karijord et al. (1982). The proportion of variance attributable to herd is smaller for C4:0 to C14:0 (approximately 0.25) than for unsaturated C18 FA (approximately 0.50). There can be many reasons for variance attributable to herd (Jensen, 2002); feed differences among farms, but also other management factors, might play a role. Possible reasons for this could be that short-chain FA are synthesized *de novo* by the cow. Long-chain FA, however, originate predominantly from dietary FA, and

because plant material contains mainly long-chain FA, differences in diet affect long-chain FA more than short-chain FA. The difference caused by herd effects, however, was less clear in the FA groups, where herd consistently explained approximately 30% of the variance. The difference between the FA was further evident from genetic correlations between FA. Strong positive genetic correlations were reported for both C6:0 to C14:0 and unsaturated C18 FA. To the contrary, genetic correlations of C6:0 to C14:0 with unsaturated C18 FA were

Table 5. Genetic correlations¹ between 4:0, 18:0, conjugated linoleic acid (CLA) *cis*-9, *trans*-11, ratio of SFA:UFA, and groups of fatty acids,² measured on a test-day morning milk sample of 1,918 cows in first lactation

Trait	C4:0	C6-12	C14-16	C18:0	C18u	CLA
C6-12	-0.36					
C14-16	0.42	-0.43				
C18:0	0.15	-0.25	0.21			
C18u	-0.17	-0.30	-0.62	-0.41		
C18:2 <i>cis</i> -9, <i>trans</i> -11 (CLA)	-0.14	-0.09	-0.60	-0.58	0.71	
Ratio ³ SFA:UFA	0.19	0.30	0.67	0.45	-0.96	-0.71

¹SE between 0.02 and 0.28.

²Groups are defined in Table 1.

³SFA = saturated fatty acids; UFA = unsaturated fatty acids.

Table 6. Genetic correlations¹ between individual fatty acids, groups of fatty acids,² and milk production traits, measured on a test-day morning milk sample of 1,918 cows in first lactation

Trait	Fat percentage	Protein percentage	Fat yield	Protein yield	Milk yield
Individual fatty acids					
C4:0	0.16	-0.11	0.30	0.04	0.09
C6:0	0.46	0.17	0.58	0.12	0.01
C8:0	0.34	0.25	0.45	0.23	0.03
C10:0	0.09	0.19	0.24	0.28	0.10
C12:0	0.00	0.20	0.15	0.25	0.09
C14:0	-0.43	-0.10	-0.11	0.33	0.30
C16:0	0.65	0.32	0.18	-0.49	-0.50
C18:0	0.01	-0.48	0.18	-0.13	0.15
C18:1 <i>cis</i> -9	-0.63	-0.32	-0.36	0.23	0.32
C18:1 <i>cis</i> -11	-0.45	-0.02	-0.17	0.46	0.29
C18:1 <i>trans</i> -4-8	-0.78	-0.35	-0.38	0.33	0.41
C18:1 <i>trans</i> -9	-0.76	-0.26	-0.29	0.57	0.54
C18:1 <i>trans</i> -11	-0.43	-0.11	-0.13	0.38	0.34
C18:2 <i>cis</i> -9,12	-0.70	-0.35	0.04	0.94	0.77
C18:2 <i>cis</i> -9, <i>trans</i> -11 (CLA ³)	-0.58	-0.02	-0.30	0.40	0.33
C18:3 <i>cis</i> -9,12,15	-0.75	-0.28	-0.28	0.60	0.53
Groups of fatty acids					
C6-12	0.14	0.23	0.26	0.24	0.06
C14-16	0.65	0.38	0.13	-0.57	-0.57
C18u	-0.72	-0.35	-0.35	0.38	0.43
Ratio ⁴ SFA:UFA	0.56	0.21	0.37	-0.19	-0.23

¹SE between 0.08 and 0.29.²Groups defined in Table 1.³CLA = conjugated linoleic acid.⁴SFA = saturated fatty acids; UFA = unsaturated fatty acids.

weak, as were genetic correlations of C4:0, C16:0, and C18:0 with the other FA. Few papers have studied genetic correlations between FA. Genetic correlations found in the current study were comparable to the results reported by Karijord et al. (1982).

Explanation for the grouping of the FA can be found in the biological pathways of synthesis. The FA C4:0 (butyric acid) was negatively correlated with almost all other FA. It is formed partly in the rumen by bacterial processes, together with acetate and propionate, and is a precursor for most other short- and medium-chain FA. Increased *de novo* synthesis will possibly convert more C4, so less C4 is present in milk, hence the negative correlation. The C6:0 to C16:0 FA are synthesized *de novo* in a FA cycle starting from C2 and C4 (Bobe et al., 1999). The C16:0, however, is partly synthesized *de novo* and is partly excreted from blood, which might explain the correlations found for this FA. The unsaturated C18 FA originate mainly from dietary FA, and their proportions are highly dependent on rumen biohydrogenation and on Δ^9 -desaturase enzymatic activity in the mammary gland (MacGibbon and Taylor, 2006).

Because of the synthesis pathways, short- and medium-chain FA are expected to be under stronger genetic control than the long-chain FA. This is also reflected in the higher heritability estimates and the smaller influence of herd for short- and medium-chain

FA, compared with long-chain FA. Genetic selection is therefore likely to be more effective for short- and medium-chain FA.

Correlation Between Fat Percentage and FA

Average milk fat percentage in the Netherlands has increased from 3.7% in 1950 to 4.4% in 2005 (NRS, 2006). A positive genetic correlation was found in the present study between fat percentage and C16:0, and a negative genetic correlation was found between fat percentage and unsaturated C18 FA. As a result of the increase in fat percentage, a correlated increase in C16:0 and a decrease in unsaturated C18 FA were expected. In 1974, Renner and Kosmack (1974a) found a fat percentage of 4%, with 25.5% C16:0 and 31.1% unsaturated C18 FA. In the current study, the fat percentage was 4.36%, with 32.6% C16:0 and 21.6% unsaturated C18 FA. Thus, comparing results from this study with those from Renner and Kosmack (1974a), fat percentage has increased, with a strong increase in the proportion of C16:0 and an even stronger decrease in the proportions of unsaturated C18 FA. Although there may be many reasons for these differences, such as breed, season, lactation stage, time of day, or feed, the change in milk fat composition could have been a corre-

lated response to selection for fat, favoring C16:0 rather than increasing all FA simultaneously.

Selection for Fat Composition

In the past, some studies questioned the need to change the milk fat composition for mainly 2 reasons: 1) because large changes in milk fat composition would be required to substantially decrease risks to human health (Maijala, 1995); and 2) because changes that are positive for one product might be detrimental for other products. The latter reason might imply that multiple breeding goals are needed and that the entire production chain would have to be adapted, for example by separately collecting milk for different end uses (Gibson, 1991). In recent years, however, there has been increased awareness among consumers and increased market demands for differentiated, specific, and healthy dairy products, even if these require large adaptations within the current production structure. Even if individual products have small effects because they are only a small part of the total diet, ultimately the sum of all improved products might result in an overall beneficial health effect, for example, by decreasing the intake of C14:0 and C16:0 FA and increasing the intake of unsaturated C18 FA. However, before a breeding program can focus on milk fat composition, several issues need to be resolved. Among others, measuring FA proportions by gas chromatography analysis is too expensive to use in routine analysis, and cheaper methods have to become available, for example, using infrared spectroscopy (Soyeurt et al., 2006b). Farmers in many countries are currently paid based on yields of fat and protein, which tend to increase rather than decrease the amounts of C14-16, so FA-based payment should possibly be introduced. Finally, the direction of selection might depend on the purpose of the milk product, because changes that are favorable for one product might be unfavorable for others. In addition to these practical considerations, it would seem worthwhile to study repeated samples from all cows to estimate parameters more accurately and to study physiological changes when selecting for or against certain FA. This study provides the necessary first data to evaluate the possibilities of improving milk fat composition by selective breeding.

CONCLUSIONS

Results have shown considerable genetic variation for all milk FA, with C4:0 to C16:0 having higher intraherd heritabilities (around 0.60) than C18 FA (around 0.25). High genetic correlations exist within the groups of short- and long-chain FA, which coincide with the

origin of the FA and the biological pathways of synthesis. Selection for fat may lead to an increased proportion of C16:0 and a decreased proportion of unsaturated C18 FA because of the high genetic correlations. This study shows that it is possible to change the milk FA composition by genetic selection.

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