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Covariance among milking frequency, milk yield, and milk composition from automatically milked cows

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ABSTRACT

Automatic milking systems allow cows voluntary access to milking and concentrates within set limits. This leads to large variation in milking intervals, both within and between cows, which further affects yield per milking and composition of milk. This study aimed to describe the degree to which differences in milking interval were attributable to individual cows, and how this correlated to individual differences in yield and composition of milk throughout lactation. Data from 288,366 milkings from 664 cow-lactations were used, of which 229,020 milkings had milk composition results. Cows were Holsteins, Red Danes, and Jerseys in parities 1, 2, and 3. Data were analyzed using a linear mixed model, with cow-lactation as a random effect and assuming heterogeneous residual variance over the lactation. Cow-lactation variance was fitted using linear spline functions with 5 knot-points. Residual variance was generally greatest in early lactation and declined thereafter. Accordingly, animal-related variance tended to increase with progression of lactation. Milking frequency (the reverse of milking interval) was found to be moderately repeatable throughout lactation. Daily milk yield expressed per milking was found to be highly repeatable in all breeds, with the highest values occurring by the end of lactation. Fat percentage had only moderate repeatability in early to mid lactation but increased toward the end of lactation. Individual level correlations showed that cows with higher milking frequency also had greater yields, but had lower fat percentage. Correlations were slightly weaker in very early lactation than in the remaining parts of lactation. We concluded that individual differences exist among cows milked automatically. Cows with higher yields are milked more often and have lower fat content in their milk.

Key words: automatic milking, repeatability, correlation, variance heterogeneity

INTRODUCTION

Automated milking allow cows the freedom to go to milking at any time and to dynamically change intervals between milkings throughout lactation as well as on a daily basis. Cows may have individual preferences for visiting the milking units more or less often. The large variation observed in milking intervals from automatically milked cows can therefore be partly attributed to systematic factors, such as stage of lactation, and to random effects between and within individual cows. Systematic manipulation of milking frequency in conventional systems is known to affect yield and composition of milk. For example, comparison between fixed milking frequencies of 2 and 3 times a day showed that milk yield was increased by up to 14% with more frequent milking (e.g., Smith et al., 2002; McNamara et al., 2008). Erdman and Varner (1995) reported that increasing the milking frequency from 2 to 3 times daily resulted in a fixed increase of 3.5 kg/d of milk yield and 92 g/d of fat yield. When milking frequency was reduced to once a day, milk yield decreased by as much as 24 to 40% (McNamara et al., 2008; Remond et al., 2009). Studies on the effect of milking frequency on milk composition have not reported consistent results. For example, Smith et al. (2002) reported that milk fat and protein percentages were significantly lower in herds milking 3 times a day than in those milking twice a day. However, Allen et al. (1986) reported only slightly lower milk fat percentage from cows milked 3 times compared with cows milked twice a day, and Amos et al. (1985) and DePeters et al. (1985) indicated that milk composition was not affected by milking frequency. The few studies that reported on once-a-day milking indicated an increase in fat content (e.g., McNamara et al., 2008; Remond et al., 2009). How changes in milking frequency between individual cows over a lactation affect milk yield and composition in automatically milked cows has not been well described.

Short-term changes to milking intervals also affect yield and composition at a given milking, but may not affect daily average yield or composition (Remond et al., 2009). For official recording purposes, moving aver-

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ages of yield over 5 to 7 d are recommended (ICAR, 2009). However, the composition of single milk samples and especially fat content shows large variation between milkings from the same cow, which can be effectively adjusted for using deviations in the preceding milking interval or yield (Friggens and Rasmussen, 2001). Other disturbances to milkings and single records come from incomplete milkings and milkings with manual intervention. To dynamically follow variation in yield and composition from milking to milking, records from “undisturbed” milkings adjusted for effects of preceding milking interval would be useful.

Only a few studies have considered variation in milking frequency caused by differences between individual cows milked voluntarily in an automated milking system (e.g., Gäde et al., 2006; Nixon et al., 2009; André et al., 2010) and the correlated effects on milk composition and yield. Individual variation in milking traits is assumed to have a genetic background (e.g., König et al., 2006) and, as such, information on individual-level covariance would mirror genetic relationships and thus be useful for dairy cattle breeding. In addition, procedures for optimizing dairy herds and use of automatic milking systems for maximizing herd or cow yield rely on information about variance components for individual cows (André et al., 2010). However, phenotypic or genetic covariance parameter estimates from automatically milked cows are sparsely reported (e.g., König et al., 2006; Nixon et al., 2009; André et al., 2010).

Estimates of variance components from conventionally milked cows show that both the residual variance and the animal-related variance change during lactation. This culminates in residual variance being greatest in early lactation and declining as lactation advances (Olori et al., 1999). In contrast, the animal or genetic variance for some traits seems to increase in later stages of lactation (Olori et al., 1999). Simple models assuming constant variance components throughout lactation are therefore insufficient and may bias the correlations between traits. However, methods for estimation of heterogeneous residual variance and functions describing variance components are available (e.g., Misztal, 2006).

The hypothesis for the current study was that the variance and relationships among milking frequency, milk yield, and milk composition in terms of fat and protein content are partly determined by characteristics of the individual cow change during lactation and may also be affected by breed of cow. Therefore, the present study aimed to test this hypothesis by studying the relationships among milking intervals, milk yield, and composition during lactation from cows of 3 dairy breeds milked voluntarily in automatic milking systems (AMS) in a research herd with intensive recording of milking traits and milk composition.

MATERIALS AND METHODS

Design and Animals

This was a cohort study of cows accessing a voluntary (automatic) milking system (VMS; DeLaval, Tumba, Sweden), carried out in the research herd at the Danish Cattle Research Centre (DCRC, Foulum, Denmark). The herd was established in September 2001, and intensive recording commenced by November 2002 and continued almost unchanged until the end of October 2006. The intensive period was used for the present study. The herd included approximately 150 cows in milk, of the Red Dane, Holstein, and Jersey breeds. Cows were primarily of first, second, and third parities because replacement priority was given to young cows. Data were restricted to the period between 5 and 305 d in milk and to cows in their first 3 parities. Cows were kept in 3 groups. Two groups consisted of Red Danes and Holsteins ($n = 52$ per group), and the third group comprised Jerseys cows ($n = 40$ cows). The Jersey group could be viewed as a separate herd because they were kept in a separate group, unlike the Red Danes and Holsteins that were housed together. Each group could access only one milking unit (VMS1, VMS2, or VMS3). The herd was housed indoors throughout the year in a freestall barn. Feeding was based on a TMR offered ad libitum. Detailed descriptions of the feed composition were reported previously (Bossen et al., 2009; Bossen and Weisbjerg, 2009). In the VMS, cows were supplied with concentrates in restricted amounts while being milked. The amount of concentrates offered at any given milking was determined from the hours since last milking. Feed allocation during milking was controlled by settings in the VMS management software (Alpro, DeLaval).

Milking Frequency, Yield, and Composition

Milkings and milk yield recordings were obtained automatically. Amounts of milk were recorded using the built in FloMaster units (DeLaval), and milk samples were collected using a modified automatic sampler (XMS, DeLaval; Løvendahl and Bjerring, 2006). Initially, milk was sampled every day for 24 h, but from April 2005, sampling intensity was reduced to five 24-h periods per week. Milk samples were preserved with Bronopol and kept cold (2°C) until analyzed for contents of fat, protein, and lactose and cell count using a CombiFoss 4000 analyzer (Foss Electric, Hillerød, Denmark) operated by the regional milk-recording society (RYK, Vojens, Denmark and Eurofins Steins Laboratorium, Holstebro, Denmark).

Cow-traffic was free; hence, cows had access to milking units at all times, except during 2 daily cleaning

Table 1. Milk recording data obtained using voluntary access to milking units by omitting data from disturbed milkings¹

Trait	Factor level	Red Dane			Holstein			Jersey		
		n	Mean/ median	SD	n	Mean/ median	SD	n	Mean/ median	SD
Lactations		267			238			159		
Calving age, d	Parity 1	124	808	62	116	802	66	69	794	68
	Parity 2	94	1,202	86	73	1,206	98	59	1,197	95
	Parity 3	49	1,564	91	49	1,588	134	31	1,613	115
Per undisturbed milking										
Milking interval, h		111,298	8.76 ²	3.32	101,931	8.86 ²	3.49	75,137	7.87 ²	2.71
Yield, kg		111,298	9.86 ²	3.94	101,931	10.88 ²	4.41	75,137	6.45 ²	2.63
Fat, %		88,913	4.32	0.81	81,054	4.14	0.79	59,053	6.32	1.04
Protein, %		88,913	3.56	0.35	81,054	3.39	0.32	59,053	4.24	0.42
Lactose, %		88,913	4.85	0.20	81,054	4.88	0.20	59,053	4.76	0.22
SCC, ln units		88,913	4.59	1.01	81,054	4.61	1.09	59,053	4.54	1.00
Per 24 h										
Milking frequency		111,298	2.74 ²	0.79	101,931	2.71 ²	0.84	75,137	3.05 ²	0.74
Milk yield, kg		111,298	26.24	7.93	101,931	28.87	8.15	75,137	19.58	5.32
Fat, kg		88,105	1.115	0.344	81,054	1.176	0.339	59,053	1.214	0.315
Protein, kg		88,105	0.919	0.242	81,054	0.967	0.246	59,053	0.820	0.195
Lactose, kg		88,105	1.275	0.393	81,054	1.412	0.409	59,053	0.937	0.266
ECM, kg		88,105	27.23	7.60	81,054	29.06	7.64	59,053	25.94	6.37

¹Undisturbed milkings = automated and complete, for both current and preceding milking.

²Medians were used for milking interval, milking frequency, and yield per milking because the traits had skewed distributions.

sessions. A minimum visit cycle was set at 4 h. Cows visiting the VMS before the set minimum time limit were rejected and were recorded as such. At the high end, cows not milked for more than 20 h were put on an attention list and fetched for milking. Cows fetched for milking were recorded manually. Automatic and manual milkings were recorded as “complete” or as “incomplete” depending on the level of success of the milking. Incomplete records occurred when the teat-cups were kicked off or failed due to any other criteria as set by the Alpro management software. Because the sequence of previous events at milking may disturb records at subsequent milkings, records were assigned to categories. For the current analysis, only records fulfilling the criterion of being “undisturbed” were used. Milkings were defined as undisturbed if they were “automated and complete” and were preceded by another automated and complete milking of the same cow, and if the cow was not fetched at the current milking. Milkings with mastitis warnings could still be classified as undisturbed. Undisturbed milkings could be preceded by one or more automated rejections, in which the cow visited the VMS without getting a food reward or being milked. The total number of undisturbed milkings was 101,931 yield records from Holstein, 111,298 from Red Dane cows, and 75,137 records from Jersey cows (Table 1).

At each milking, the identity of the cow was recorded and used to trace information on DIM, parity,

and breed. Through date and clock time, the intervals since previous milkings were calculated for any type of milking or visit (completed, uncompleted, fetched, or rejected). Milking interval was expressed in decimal hours and as milking frequency per 24 h. From each milking, the yield in kilograms and the milk contents of fat, protein, lactose, and somatic cells were used to calculate amounts of fat, protein, and lactose in kilograms per milking, and as yield per 24 h. Milk yield records of less than 0.2 kg were considered false milkings, and yields were excluded from further analysis. The yield per 24 h was calculated as an extrapolation of the yield per milking using the following curvilinear equation:

$$\text{Yield}_{24} = -0.1745 + 0.6213 \times \text{MilkFreq} + \text{MilkYield} \times (0.2806 + 0.8118 \times \text{MilkFreq}),$$

where Yield₂₄ is the calculated milk yield per 24 h; MilkFreq is the milking frequency for the current milking; and MilkYield is the yield recorded at the current milking. The formula was obtained by linear regression of a 5-d moving average yield per 24 h as suggested by ICAR (2009) on yield per milking and milking frequency. This formula takes into account that milk production per hour is only almost constant but will decline with longer milking intervals (e.g., Remond et al., 2009). This general equation was used for cows of all 3 breeds and parities throughout lactation. By cal-

culating yield per 24 h this way for every undisturbed milking, cows would have this variable expressed repeatedly within each day.

Fat content of milk (Fat_b) is affected systematically by deviations in milking interval or yield from the given cow's moving average interval or yield (Friggens and Rasmussen, 2001). Preadjustment of fat percentage for deviations in milking interval was performed using the approach of Friggens and Rasmussen (2001) for each breed. The preadjustment coefficients were also corrected for stage of lactation to give the equations listed below (Adj_fat_b). Milking intervals were preferred to yield as the basis for adjustments to avoid circularity in formulas. This was because milking intervals were more alike across breeds and lactation stages than milk yield. Deviations in milking interval (Dev_MI) were obtained as differences between the actual record and an exponentially smoothed value for each cow ($\alpha = 0.25$) working as a weighted moving average:

$$\begin{aligned} \text{Red Dane: Adj_fat_b} &= \text{Fat_b} \\ &- (0.1505 + 0.000146 \times \text{DIM}) \times \text{Dev_MI}, \end{aligned}$$

$$\begin{aligned} \text{Holstein: Adj_fat_b} &= \text{Fat_b} - \\ &(0.1255 + 0.000144 \times \text{DIM}) \times \text{Dev_MI}, \end{aligned}$$

$$\begin{aligned} \text{Jersey: Adj_fat_b} &= \text{Fat_b} - \\ &(0.1995 + 0.000139 \times \text{DIM}) \times \text{Dev_MI}. \end{aligned}$$

Yields of fat (kg) per 24 h and of ECM (Sjaunja et al., 1990) were calculated using adjusted fat content.

Data Analysis

Linear mixed models were fitted where the random animal part was modeled using linear splines in a random regression approach (Miształ, 2006). Fixed effects included lactation curves and terms accounting for diurnal variation and calving age, systematic seasonal changes, and categorical effects of milking unit and parity. Systematic effects of diets (TMR rations) experimentally assigned to groups of cows were included as fixed effects. The models had the general form

$$\mathbf{Y} = \mathbf{Xa} + \mathbf{Zu} + \mathbf{Qd} + \mathbf{Se}, \quad [1]$$

where \mathbf{Y} is the vector of observed response (e.g., milking frequency, milk yield, fat percentage). Fixed (systematic factors) effects are in vector \mathbf{a} with design or incidence matrix \mathbf{X} . Random animal effects are in vector \mathbf{u} , with incidence matrix \mathbf{Z} , and test-day "noise" is in \mathbf{d} with

incidence matrix \mathbf{Q} , and the residuals assumed to be normally distributed are in \mathbf{e} , with incidence matrix \mathbf{S} for segments of the lactation (see below). If the whole lactation is modeled as one segment, \mathbf{S} becomes an identity matrix, \mathbf{I} .

The fixed part of the model was developed and tested for one trait at a time using the MIXED procedure of SAS (version 9.2, SAS Institute Inc., Cary, NC). In this development stage, the random terms were reduced in order for the MIXED procedure to cope with computational constraints. This was mainly achieved by omitting the test-day effect (\mathbf{Qd}), thereby including it in the residual term. Lactation curves were fitted using the Wilmink function [2] (Wilmink, 1987):

$$y_t = a + be^{-kt} + ct + dt^2, \quad [2]$$

where y_t is the yield at t DIM, and a , b , c , and d are regression coefficients. The parameters jointly describe the curve shape so that a sets the amplitude of daily yield, b is the acceleration in early lactation, c the linear decline after peak, and d is the inflexion deviation over the later stages of the lactation. The fifth parameter k affects the duration of the acceleration period and was fixed at $k = 0.05$, as used by Wilmink (1987). As age (parity) and breed are known to affect lactation curve shapes (e.g., Wilmink, 1987; Hansen et al., 2006), curves were modeled within these factors.

Diurnal changes were modeled within parity using a Fourier series approach [3] previously used by Løvendahl and Bjerring (2006).

$$y_\theta = \sum_{j=1}^4 (\cos j\theta 2\pi + \sin j\theta 2\pi), \quad [3]$$

where θ is the decimal fraction of the 24-h diurnal cycle when the milking was initiated (i.e., $\theta = h/24$) and π is expressed as an angle in radians. The series was restricted to include the first $j = 4$ "harmonics." The Fourier series were tested against a model in which the diurnal rhythm was a factor with 24 levels.

The model was run within breed, although the Red Dane and Holsteins were housed together and the Jerseys kept and milked in a separate group (VMS3). For the Red Dane and Holstein, the actual group (VMS1 or VMS2) was included as a factor. Parity was fitted with 3 levels, and calving age in first lactation was fitted as a covariate. Effects of pregnancy were modeled using a linear regression on days pregnant beyond 140 d, as suggested by Strandberg and Lundberg (1991). Specific regressions were used for each parity.

The feeding regimen was fitted as 3 fixed factors, one for the type of TMR (4 levels), one for the concentrate

type (2 levels), and one for the concentrates amount (10 levels). As the feeding regimen changed during lactation, the actual factor levels were assigned to each milking. A seasonal effect of calving-month-year was initially fitted as a fixed effect. Seasonal effects were in the final model fitted as an overall sinusoidal waveform and as an interaction with each year using the Fourier approach described above but restricted to $j = 1$.

Random animal effects were defined with cow-parity as the repeated unit. Although some cows had data in more than one parity, any possible correlations between consecutive lactations were ignored because the study was focused on within-parity covariance. Covariance was modeled using random regression based on linear spline functions (Miszta, 2006). Knot points were placed at 5, 50, 110, 215, and 305 DIM, and a curvature modifier $q = 0.75$ was used to alleviate "chain lines" between knots.

To investigate the change in residual variance and other variance components during lactation, the trajectory (5 to 305 DIM) was divided into 19 segments of equal duration of 15 d and a last segment of 16 d, and allowing for residual variance to differ between segments. Residual correlations between segments were ignored, as they were assumed nonexistent. Estimates of repeatability were obtained from variance component estimates at knot-points as $t = \sigma_{cow}^2 / (\sigma_{cow}^2 + \sigma_e^2)$. Standard errors of parameter estimates were obtained from standard errors on covariance components using a Taylor series expansion. Individual level correlations between traits were obtained by using random animal estimates at knot-points for all traits. Individual-level correlated effects of higher milking frequency were also estimated as linear regressions of liquid milk, ECM, and fat content on milking frequency using random animal solutions.

RESULTS

Data from 664 cow-lactations with 288,366 milkings categorized as undisturbed were included in the study. Of these, 229,020 (79%) had milk composition information. Descriptive statistics are presented in Table 1. Milking intervals were longer in the Holstein and Red Dane cows accessing VMS 1 and 2 than in the Jersey-group (VMS 3). Similar differences were seen for the milking frequency, so that cows in the Jersey-group had more milkings per day than Holsteins and Red Danes. Yield per milking was higher in Holstein and Red Dane cows than in Jerseys. However, the concentration of milk solids (fat and protein) was lower in Holstein and Red Dane cows than in Jerseys. When expressed per 24 h, production of fat (in kg) was similar among the breeds but protein and lactose production was lowest

for Jerseys. Jointly, the production of ECM per 24 h was highest in Holsteins followed by Red Danes and Jerseys. However, the effects of the Jersey group were indistinguishable from the effect of these cows, being in a separate and smaller group than the Red Dane plus Holstein groups.

Systematic Effects

Diurnal profiles for milking interval and other milking variables were modeled effectively using the Fourier approach, because it gave no increase to the residual variance but used fewer degrees of freedom compared with a model using 24 discrete hour classes (data not shown).

The frequency of milkings per day increased from the start of lactation to reach a peak either very early or before 2 mo, and then declined steadily as lactation progressed (Figure 1). The yield per milking and yield per day initially increased to reach a peak at 42 to 57 DIM, followed by a steady decline (not shown). The fat content was initially high but declined with increasing DIM to reach a nadir at 57 DIM, followed by a steady incline for the rest of the lactation. The yield of ECM per day peaked at 27 DIM and declined thereafter (not shown). The SCC of milk was initially high and declined relatively rapidly to reach a nadir at 27 DIM followed by a steady incline (not shown). Similar shapes of curves were found in all breeds and all lactations. Lactation curves for ECM (kg/d), milk fat content (%), and SCC (log-e units/mL) for Red Danes, Holstein, and Jersey cows were fitted using the Wilink function for parities 1 to 3 (not shown). A linear function was included to account for pregnancy effect. In all 3 lactations, Holstein cows had the highest ECM yield and Jersey cows had the lowest ECM yield. In general, first-lactation animals had the lowest milk yield, whereas parity 3 animals had the highest milk yield.

Year-season effects, modeled as first-order Fourier functions including a year-season interaction, were found to be significant for all variables ($P < 0.05$). Feeding regimen, which was included in the model to account for its effect on the dependent variables, had a significant effect on the majority of the variables. All traits were affected by the TMR type ($P < 0.001$) except SCC in Jersey cows. The concentrate strategy and amounts did not affect fat and protein concentrations or amounts of fat and protein per milking but affected all the other traits significantly ($P < 0.001$).

Variance Components and Repeatability

Estimates of random variance components showed that both animal and residual variance change over the

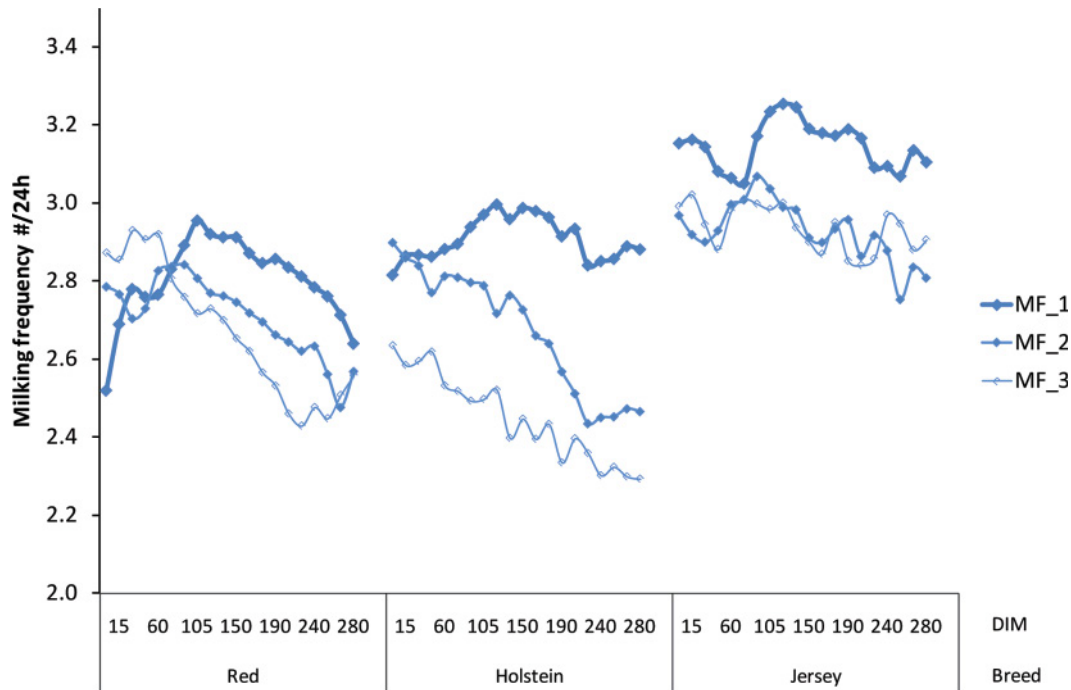


Figure 1. Milkings per day in Red Dane, Holstein, and Jersey cows. Results are means per 15-d segment in parities 1, 2, and 3. MF_x = milking frequency in parity x. Color version available in the online PDF.

lactation. However, the residual and animal variance components were more stable over the lactation for milking frequency (Figure 2) than for yield traits (i.e., ECM₂₄; Figure 3). The results showed that for some traits the repeatability was constant over the lactation, whereas for other traits the repeatability increased by the end of lactation (Table 2). Estimates of repeatability in knot-points during lactation for each breed are presented in Table 2. The changes in repeatability estimates over lactation showed similar trends for the Jersey group as for the other 2 breeds, although this group contained fewer cows.

Repeatability estimates were larger for yield per 24 h than for yield per milking throughout lactation. Variation in fat content was affected by stage of lactation. Repeatability declined from its starting value to a nadir at DIM 50 and increased thereafter. Protein and lactose contents of milk were less variable than fat content through lactation (Table 2), as was reflected in large repeatability coefficients. The repeatability of SCC was generally high and showed little change during lactation (Table 2).

Individual-Level Correlation

Correlation coefficients among variables were estimated at the level of individual cows within lactation by correlating random solutions obtained in

each knot-point from each single-trait analysis, and thereby illustrate changes in correlation over lactation (Table 3). The correlation between milking frequency and milk yield per 24 h and ECM per 24 h was positive, whereas the correlation between milking frequency and yield per milking was negative. Correlation between milking frequency and composition was negative for fat percentage and protein percentage in the Holsteins and Red Danes, but close to zero in the Jersey group. Correlation coefficients in the very early part of lactation were generally weaker than those in mid and late lactation.

Correlations between yield per milking and yield per day were close to 0.60 throughout lactation in all 3 breeds (Table 3), whereas the correlation with milking frequency was around 0.40. Correlations between yield per milking and contents of fat and protein were negative and weak ($r = -0.12$ to -0.25) in all breed groups. Individual-level correlations between fat content of milk and other milk variables were weak at the start of lactation and stable from DIM 50 onward, except for the moderate to strong correlation with protein percentage. Energy-corrected milk is a weighted expression of yield and content. The yield of ECM per day was strongly correlated with the yield in kilograms per day, but less so with milking frequency and yield per milking. The correlation between milking frequency and SCC was negative and of moderate magnitude.

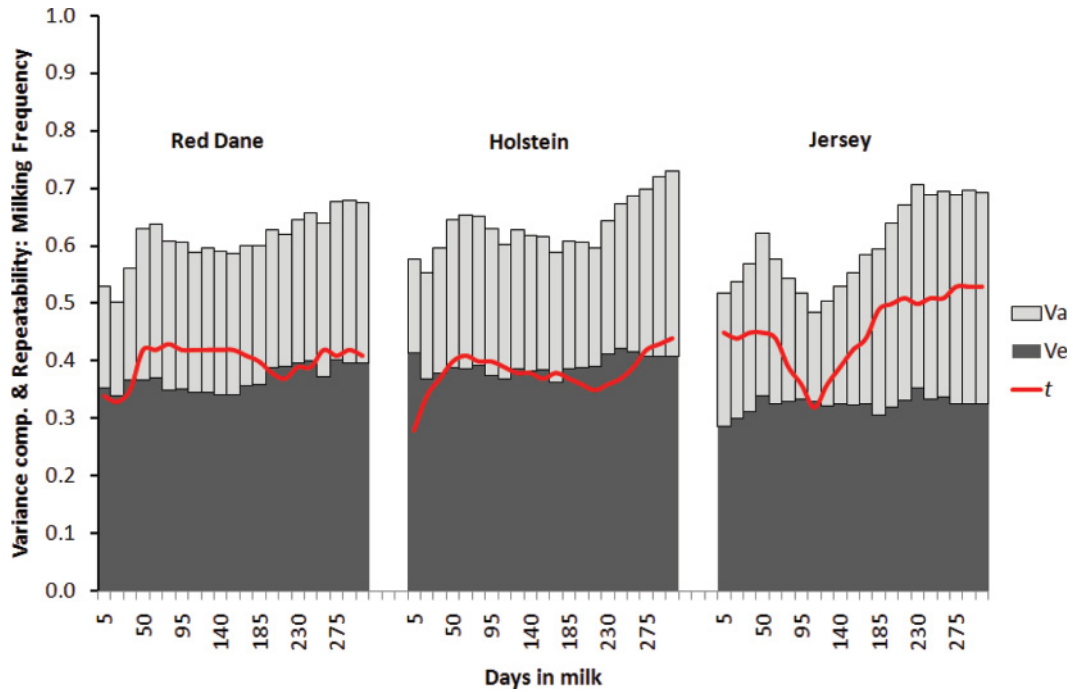


Figure 2. Estimates of residual variance in 15-d segment and cow-within-parity variance and the resulting repeatability for milking frequency in automatically milked Red Dane, Holstein, and Jersey cows. Cow-related variance was estimated using linear splines with knot points at 5, 50, 110, 215, and 305 DIM. Va = animal variance; Ve = residual variance; t = repeatability. Color version available in the online PDF.



Figure 3. Estimates of residual variance in 15-d segments and cow-within-parity variance and the resulting repeatability for daily yield of ECM in automatically milked Red Dane, Holstein, and Jersey cows. Cow-related variance was estimated using linear splines with knot points at 5, 50, 110, 215, and 305 DIM. Va = animal variance; Ve = residual variance; t = repeatability. Color version available in the online PDF.

Table 2. Estimates of repeatability for milking and milk yield traits obtained in automatic milking system for Holstein, Red Dane, and Jersey cows in the first 3 parities at 5, 50, 110, 215, and 305 DIM

Item	Breed	Repeatability ¹				
		5	50	110	215	305
Milking frequency	Red Dane	0.34	0.42	0.42	0.37	0.41
	Holstein	0.28	0.40	0.39	0.35	0.44
	Jersey	0.45	0.45	0.32	0.51	0.53
Yield, kg/milking	Red Dane	0.54	0.56	0.51	0.55	0.61
	Holstein	0.52	0.55	0.52	0.53	0.66
	Jersey	0.64	0.52	0.44	0.68	0.72
Yield, kg/d	Red Dane	0.74	0.81	0.84	0.88	0.92
	Holstein	0.82	0.84	0.84	0.84	0.93
	Jersey	0.77	0.70	0.76	0.87	0.92
Fat, %	Red Dane	0.43	0.39	0.48	0.50	0.65
	Holstein	0.63	0.45	0.47	0.59	0.63
	Jersey	0.66	0.31	0.41	0.47	0.48
Protein, %	Red Dane	0.73	0.81	0.81	0.84	0.84
	Holstein	0.82	0.79	0.82	0.81	0.84
	Jersey	0.70	0.80	0.67	0.83	0.57
Yield fat + protein, kg/d	Red Dane	0.67	0.60	0.70	0.85	0.89
	Holstein	0.80	0.70	0.74	0.81	0.90
	Jersey	0.63	0.49	0.57	0.77	0.83
Yield ECM, kg/d	Red Dane	0.69	0.64	0.73	0.86	0.90
	Holstein	0.81	0.73	0.76	0.82	0.91
	Jersey	0.64	0.50	0.58	0.78	0.84
Somatic cells, log-e	Red Dane	0.77	0.72	0.70	0.77	0.79
	Holstein	0.73	0.70	0.72	0.76	0.78
	Jersey	0.69	0.74	0.79	0.77	0.77

¹Standard error of repeatability estimates was <0.03 for Red Dane and Holstein and 0.04 for Jersey.

Cows with higher milking frequency gave more liquid milk per day, amounting to a 20% increase in Red Dane and Holsteins and 16% in Jersey cows. The higher milking frequency was followed by decreased fat content with -0.29 percentage units in Red Dane, -0.16 percentage units in Holsteins, and -0.17 percentage units in Jerseys. The joint effects on yield as ECM were increases of 4.62 (± 0.33), 5.31 (± 0.37), and 3.61 (± 0.31) kg/d in Red Dane, Holstein, and Jersey cows. Changes tended to be larger in parities 2 and 3 than in first parity, and changes tended to be larger in late than in early lactation (results not shown).

DISCUSSION

Overall

The results of the present study have provided estimates of individual and residual variance for milking frequency for cows milked voluntarily in an AMS. Further, the results have shown that variation in milking frequency is an attribute of the individual cow, and that random residual variance remains large and

stable throughout lactation. Milk yield per milking was moderately repeatable throughout lactation, although individual and residual variance components changed during lactation. Yield per 24 h, expressed as a calculated measure at every milking following adjustment for preceding milking interval, was more highly repeatable than yield per milking throughout lactation. The correlations among milking interval, milk yield per milking and per 24 h, and fat and protein contents showed that cows with higher yields gave more milk per milking and were milked more often, but gave milk with lower fat content. However, when yield was calculated as ECM, the correlation to fat content became close to zero. The individual correlations were constant across the lactation stages except for the start of lactation and were in most cases similar across breed groups.

Expression of Yield and Adjustments for Systematic Effects

The statistical model included several systematic factors, primarily to isolate their effects from the random variance components. Before running the models,

Table 3. Individual level correlations between milking traits at 5, 50, 110, 215, and 305 DIM¹

Trait/breed	Milking frequency					Yield, kg/milking					Fat content, %					ECM, kg/d				
	5	50	110	215	305	5	50	110	215	305	5	50	110	215	305	5	50	110	215	305
Yield, kg/milking																				
Red Dane	-0.47	-0.46	-0.41	-0.21	-0.15						-0.10	-0.14	-0.23	-0.06	-0.07	0.65	0.57	0.56	0.70	0.71
Holstein	-0.34	-0.36	-0.38	-0.31	-0.28						-0.17	-0.24	-0.21	-0.24	-0.14	0.67	0.59	0.57	0.63	0.67
Jersey	-0.68	-0.58	-0.48	-0.47	-0.15						-0.20	-0.22	-0.34	-0.26	-0.36	0.46	0.41	0.50	0.49	0.58
All	-0.46	-0.44	-0.39	-0.30	-0.20						-0.12	-0.19	-0.22	-0.16	-0.15	0.63	0.56	0.56	0.64	0.66
Yield, kg/d																				
Red Dane	0.22	0.39	0.41	0.44	0.48	0.69	0.56	0.58	0.72	0.69	-0.17	-0.34	-0.40	-0.24	-0.32	0.91	0.90	0.90	0.94	0.95
Holstein	0.32	0.41	0.41	0.41	0.43	0.72	0.65	0.64	0.66	0.66	-0.31	-0.33	-0.29	-0.32	-0.26	0.88	0.90	0.92	0.92	0.95
Jersey	0.09	0.39	0.32	0.43	0.54	0.61	0.47	0.60	0.52	0.63	-0.34	-0.46	-0.48	-0.42	-0.48	0.76	0.86	0.85	0.91	0.92
All	0.22	0.37	0.39	0.41	0.47	0.69	0.60	0.62	0.66	0.66	-0.23	-0.34	-0.34	-0.31	-0.32	0.88	0.89	0.90	0.93	0.94
Fat, %																				
Red Dane	-0.11	-0.22	-0.23	-0.22	-0.38	-0.10	-0.14	-0.23	-0.06	-0.07						0.12	-0.00	-0.11	0.01	-0.11
Holstein	-0.22	-0.13	-0.07	-0.10	-0.15	-0.17	-0.24	-0.21	-0.24	-0.14						-0.04	-0.01	-0.03	-0.02	-0.03
Jersey	-0.03	-0.20	-0.09	-0.12	-0.31	-0.20	-0.22	-0.34	-0.26	-0.36						0.13	-0.12	-0.12	-0.19	-0.28
All	-0.13	-0.18	-0.13	-0.16	-0.28	-0.12	-0.19	-0.22	-0.16	-0.15						0.06	-0.03	-0.07	-0.05	-0.12
Protein, %																				
Red Dane	-0.14	-0.27	-0.26	-0.33	-0.38	-0.15	-0.19	-0.25	-0.23	-0.20	0.12	0.43	0.55	0.53	0.52	-0.19	-0.24	-0.25	-0.29	-0.30
Holstein	-0.13	-0.22	-0.16	-0.29	-0.22	-0.03	-0.19	-0.23	-0.23	-0.28	-0.10	0.38	0.57	0.54	0.57	-0.09	-0.17	-0.19	-0.23	-0.26
Jersey	0.22	-0.07	0.08	0.07	0.06	-0.25	-0.19	-0.34	-0.32	-0.24	0.15	0.41	0.63	0.70	0.57	0.08	-0.07	0.02	-0.10	0.02
All	-0.04	-0.21	-0.15	-0.18	-0.22	-0.12	-0.16	-0.24	-0.25	-0.23	0.04	0.40	0.58	0.57	0.53	-0.11	-0.17	-0.17	-0.22	-0.23
ECM, kg/d																				
Red Dane	0.19	0.33	0.39	0.43	0.45	0.65	0.57	0.56	0.70	0.71	0.12	-0.00	-0.11	0.01	-0.11					
Holstein	0.31	0.41	0.43	0.40	0.40	0.67	0.59	0.57	0.63	0.67	-0.04	-0.01	-0.03	-0.02	-0.03					
Jersey	0.11	0.36	0.36	0.44	0.53	0.46	0.41	0.50	0.49	0.58	0.13	-0.12	-0.12	-0.19	-0.28					
All	0.21	0.36	0.40	0.41	0.45	0.63	0.56	0.56	0.64	0.66	0.06	-0.03	-0.07	-0.05	-0.12					
Log SCC																				
Red Dane	-0.09	-0.16	-0.24	-0.31	-0.24	-0.15	-0.15	-0.08	-0.25	-0.25	0.02	0.07	0.07	0.04	0.19	-0.25	-0.33	-0.30	-0.46	-0.40
Holstein	-0.18	-0.14	-0.29	-0.15	-0.19	-0.07	0.01	-0.01	-0.14	-0.20	0.15	0.13	0.16	0.05	0.09	-0.18	-0.11	-0.24	-0.28	-0.32
Jersey	-0.09	-0.24	-0.31	-0.31	-0.33	-0.08	0.18	0.12	-0.06	-0.07	0.00	-0.16	-0.11	-0.05	0.12	-0.18	-0.14	-0.29	-0.41	-0.37
All	-0.12	-0.18	-0.27	-0.25	-0.25	-0.11	-0.01	-0.01	-0.16	-0.19	0.07	0.04	0.06	0.02	0.13	-0.21	-0.19	-0.27	-0.38	-0.37

¹Estimates were obtained as correlations between random animal solutions in 5 knot-points placed at 5, 50, 110, 215, and 305 DIM. Separate estimates were obtained for each breed (Red Dane, Holstein, and Jersey) and a pooled estimate across breeds. Correlations were significant at $P < 0.05$ if they numerically exceeded 0.12 (Red Dane), 0.13 (Holstein), 0.16 (Jersey), and 0.08 (All).

data were filtered to remove extremely outlying observations, and only milkings defined as undisturbed fulfilled the criterion of being voluntary and were used for estimation of variance components. This procedure caused a loss of data, but the data lost were considered to contain more “noise” than information because of possible disturbances before or during the milking procedure. Milk yield data were recorded at every milking, but were highly variable and dependent on the preceding milking interval. A curvilinear function using the current milk yield and the preceding milking interval was used to calculate milk yield on a 24-h basis at every milking. The function took into account that secretion rate of milk was dependent on the preceding milking interval in a declining way (e.g., Remond et al., 2009). The advantage of this approach was that dynamic changes could be followed closely in time. The disadvantage was that the measurement error affected every single observation (the residual variance). Fat content was also influenced by preceding milking interval, and adjustments using deviations in milking intervals were done applying the concept of Friggens and Rasmussen (2001). Differences between morning and afternoon yield and composition in conventional milking systems are well known. However, automatic milkings are recorded continuously, without fixed intervals. The effects of having no definite fixed milking intervals on milk yield and composition changes were accounted for by modeling them as waveforms using Fourier functions of the time of day (Løvendahl and Bjerring, 2006). Similarly, seasonal effects were modeled using the Fourier approach, within calendar year.

Residual Variance

The current study found heterogeneous residual variance by segmenting lactations into 15-d periods and by including the segments as additional random structures in the model. Other studies of lactation data have detected heterogeneity in residual variance using segments (e.g., Olori et al., 1999). By allowing for heterogeneous residual variance, estimates of other variance components become unbiased. This resulted in more reliable repeatability estimates. Obviously, the number of segments needed depends on the differences in variances between segments. Reducing the number to 4 was found sufficient for genetic analysis of yield in the study by Olori et al. (1999).

Repeatability

Linear splines in random regression models were used in the current study to estimate individual animal variance as a single cow-within-lactation component

and to express it as repeatability within breed groups. Milking frequency had intermediate repeatability of around 0.40. Repeatability values were slightly higher by the end of lactation. Similar estimates of repeatability for milking frequency were obtained by Nixon et al. (2009) and slightly lower estimates were found by König et al. (2006). These findings show that milking frequency is a repeatable trait, and the literature points to individual variation being partly caused by genetic differences (König et al., 2006). Yield expressed at every milking on a per 24-h basis had greater repeatability than was found for yield per milking, showing that the adjustments were effective. This way, repeatability for yield per day expressed at every milking was similar to estimates reported by Nixon et al. (2009) for yield based on the sum of milkings over a 24-h period, and to repeatability for yield per day in a twice-a-day system (Løvendahl et al., 2003). Fat percentage was found to be the least repeatable trait even after adjustments for deviations in milking interval. Repeatability of composition traits in single milk samples obtained in a twice-a-day milking system (Løvendahl et al., 2003) showed fat percentage to have a repeatability of 0.5 at 50 DIM, increasing to 0.7 at the end of lactation, being slightly higher than in the present study. For protein, the repeatability was around 0.8 throughout the lactation, similar to previous findings in a twice-a-day milking system (Løvendahl et al., 2003). Energy-corrected milk was intermediately to highly repeatable, with the highest repeatability by the end of lactation. This is in agreement with previous findings in cows milked twice a day (Løvendahl et al., 2003). These findings indicate that both yield and composition of milk samples (following appropriate adjustments for milking frequency) are repeatable traits, to almost the same degree in an AMS as in a twice-a-day milking system.

Correlations Estimated from Random Solutions

Within breed, individual-level correlations between random effect solutions at knot-points showed that daily milk yield and daily ECM were positively correlated to milking frequency, although less so immediately after calving. Positive correlations between yield and milking frequencies were previously reported both as genetic and as permanent environmental correlations obtained in AMS herds (König et al., 2006; Nixon et al., 2009). Correlations between yield per milking or per day with fat percentage were both negative, meaning that cows with greater yields of liquid milk gave milk that was lower in fat. Milk yield per day and ECM yield per day were highly correlated; however, ECM yield per day was almost uncorrelated to fat percentage. The individual correlations found within breeds were of similar

magnitude in the 3 breed groups, as were the patterns of change over lactation, indicating that the findings are of general nature. The associations between milking frequency and other milking traits were also quantified as correlated effects. The linear regressions showed that one more milking gave 16 to 20% more liquid milk that had less fat, resulting in a substantial increase in daily ECM production. In comparison, a 14% increase was found when going from 2 to 3 times a day milking (e.g., Smith et al., 2002; McNamara et al., 2008), or as fixed units of 3.5 kg/d (Erdman and Varner, 1995). A reduction in milking frequency from twice to once per day reduced yield by up to 40% (McNamara et al., 2008; Remond et al., 2009). Effects of systematically increased milking frequency on milk composition have been less consistent, but mostly indicate a reduction in fat content with higher milking frequency (e.g., DePeters et al., 1985; Allen et al., 1986; Smith et al., 2002). Thus, the associations based on individual animal differences seem to closely mirror the effects of systematic changes. It is also tempting to speculate that the increase in yield with higher milking frequency is mediated through counteracting the “roll-off” effect of long milking intervals. These results were based on intensive records from a research herd. The specific effects of the Jersey breed could not be separated from effects of smaller group size or less occupancy rate of the milking unit. Nonetheless, the results have shown good consistency across breed-groups indicating that findings may also hold in other herds. This entails that future cows will cope with higher yields, at least partly, by increasing milking frequency. Therefore, the demands of future generations of cows need to be taken into account when optimizing capacity and settings of the AMS (André et al., 2010). Priorities may differ, however, between herd managers. If priority is given to obtaining the highest yield per cow, the correlations obtained in this study suggest that cows will express the highest yields when they are allowed to have a high milking frequency. It is tempting to assume that the higher milking frequency found with higher yield is caused by increased appetite, as Prescott et al. (1998) showed that the food reward given in the AMS was the main motivation for visits. Correlations between traits did not approach unity. This shows that cows may still express high yields even when restricted in milking frequency, as they have traditionally been in twice-a-day milking systems. In that case, each milking may take longer or cows with a higher milk flow rate may be preferred (Gäde et al., 2006). Before extrapolating, we must also assume that individual cow differences have a genetic background, so that further studies on the correlated response of other traits to selection for high milk yield for cows in AMS would be valuable to

breeding as well as management decisions. Such investigations should include a wider range of milking traits, including milkability or flow rate.

CONCLUSIONS

The current study provides evidence for a strong individual variation in milking interval that correlates to individual variation in milk yield and milk composition in terms of fat and protein content. The correlations mean that cows with higher yield achieve their higher yields by combining higher yield per milking with higher milking frequency when allowed almost free access to milking units. We found evidence for change in variance components during lactation, but correlations between milking traits appear to be stable except during the first part of lactation. Repeatability and correlation coefficients tended to be similar for Holstein and Red Dane cows, and only slightly different for Jersey cows, although Jersey cows were kept in a smaller group, allowing less occupation of the milking unit.

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