Association of individual cow milk fatty acid proportion and variance with milk production

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Abstract

Milk fatty acid (FA) fractions, de novo (DN), mixed, and preformed (PF), are grouped based on chain length, as < C16:0, C16:0, and > C16:0, respectively. These groups reflect the origin of milk FA synthesis, with DN representing FA synthesis within the mammary gland, PF representing FA coming from the diet or body tissue reserves and mixed representing all three sources. Understanding the associations of milk FA groups with milk and component yields at the individual cow level may provide insight into making management and dietary decisions. To investigate these associations, milk samples (n = 14,091) were collected during the morning milkings from 1,737 Holstein cows from a herd milking 3x daily and averaging 41 kgs milk/cow. Milk samples were analyzed for FA groups (g/100g fat), fat, true protein, and lactose. Time periods of the first test (FT; 30 ± 2 DIM), peak milk (PT; 68 ± 31 DIM), and mid-lactation (MT; 100 ± 2 DIM) were selected. The variance of the FA groups was calculated for each animal as the variance of FA proportion between tests days within the first 305 DIM. Linear models were fit with FA group (proportion or variance), parity (1 vs ≥ 2), their interaction, and DIM (FA proportion models only) as the fixed effects and the month of sampling (FA proportion models) or month of calving (variance models) as the random effect. Across all periods, PF was positively associated with test day milk yield and cumulative milk yield through 305 DIM. In contrast, DN was negatively associated with test day milk yield and cumulative milk yield through 305 DIM across all periods ($P < 0.1$). Interestingly, increased variation in DN within cows across the first 305 DIM was positively associated with cumulative milk yields through 305 DIM ($P< 0.01$). The relationship between FA and component yields differed among the periods. Energy-corrected milk yield had a significantly negative association with DN at FT ($P = 0.04$) and a highly significantly positive association with DN at MT ($P < 0.01$). Still, it was not significantly associated with DN at PT ($P = 0.24$). The fat yield was negatively associated with DN at FT but was positively associated with DN at PT and MT ($P \leq 0.01$). In contrast, fat yield was positively associated with PF at FT and negatively associated with PF at PT and MT ($P \leq 0.03$). Protein yield was positively associated with DN for multiparous cows, negatively associated with DN for primiparous cows at FT2 ($P < 0.01$), and positively associated with DN for all parities at MT ($P < 0.01$). In contrast, protein yield was negatively associated with PF for multiparous cows, positively associated with PF for primiparous cows at FT ($P = 0.02$), and not significantly associated with PF at PT and MT ($P > 0.1$). The association of milk FA groups with milk and component yields suggests that milk FA groups may be a useful management tool for making pen grouping decisions, cow selection and breeding decisions, and informing dietary adjustments. However, the variable associations between FA groups and milk performance outcomes by parity and at different lactation stages highlight the

importance of considering these factors when making decisions based on a single milk test. Routine milk testing across lactation may allow for tailored management decisions at the individual cow level using these FA groups.

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Introduction

Of the 9.4 million dairy cows in the United States, 41.9% are on routine Dairy Herd Improvement (**DHI**) testing (CDCB, 2024). When deciding to participate in DHI testing, the producer must consider not only the cost of testing, but also the value of the data they receive. Routine DHI testing typically provides milk yield and major component (fat, protein, lactose, SCC) information back to the producer, but there is considerably more information that can be extracted from the same milk sample using the existing technology. Of these additional traits, milk fatty acid origin groups are already available on most milk analysis instruments and have been of recent interest (Dorea and Armentano, 2017; Woolpert *et al*., 2017; 2018). Milk fatty acid origin groups, *de novo*, mixed, and preformed, are grouped based on chain length, as < C16:0, C16:0, and > C16:0, respectively. These groups reflect the origin of milk fatty acid synthesis. De novo represents fatty acid synthesis within the mammary gland, preformed represents fatty acids coming from the diet or body tissue reserves, and mixed represents all three sources. At the herd level using bulk tank milk, these fatty acid groups can be informative when making dietary and management decisions (Dorea and Armentano, 2017; Woolpert *et al*., 2017; 2018). However, less is understood about these fatty acids at the individual cow level. Understanding the associations of milk fatty acid groups with milk and component yields at the individual cow level may provide insight into making management and dietary decisions. Therefore, our objective was to identify the association of milk de novo and preformed fatty acids with milk and milk component production at different stages of lactation and investigate within-cow variability of fatty acids across lactation.

Materials and methods

Data collection

Data were collected from a privately-owned Holstein dairy in New York, USA. Milk samples were collected for 18 weeks between May and September of 2023 from all three milkings within the day. Milk samples were analysed on the same Milkoscan 7 RM instrument for milk fat, true protein, lactose, de novo fatty acids, mixed fatty acids, and preformed fatty acids (FOSS Analytical, Hilleroed, Denmark). Milk yields were recorded electronically using electronic meters (SmartControl meter, BouMatic).

Data editing

The raw dataset comprised of 63,312 milk samples from 2,687 cows. Records were removed when > 305 DIM or < 2 DIM, component records were > 3 standard deviations from the mean within milking session, and when the daily record of 3 milk yields and component values were incomplete. The cleaned dataset consisted of 42,273 records representing 14,091 daily records from 1,747 cows.

Milk fatty acids (g/100g milk) were calculated on a fat basis as:

$$\frac{fatty\ acid\ (g/\ 100g\ milk)}{milk\ fat\% \times 0.95} \times 100 = fatty\ acid\ (g/\ 100g\ fat) \quad (1)$$

effectively calculating the milk fatty acid groups as a proportion of milk fat.

In order to investigate the associations at specific stages of lactation, the data were categorized as first test (**FT**; 30 ± 2 DIM), peak milk (**PT**; 68 ± 31 DIM), and mid-lactation (**MT**; 100 ± 2 DIM). Peak test represents the milk test nearest in time to the individual cow's actual peak milk production. The first morning milk sample was used for fatty acid proportions to better reflect normal DHI test day data.

Statistical analysis

Data analysis was conducted using the lmer package of R 4.3.1 (R Core Team, 2024). For each production outcome, the model contained the fixed effects of fatty acid proportion (de novo, mixed, or preformed), parity (primiparous or multiparous), their interaction, day in milk, and the random effect of month of sampling.

Individual cow variation in fatty acid proportion across lactation was calculated in two steps. First, a Wilmink curve was fit for each cow using all fatty acid data and the deviance was determined as the difference between the observed and predicted values. Second, the variance was calculated as the log variance of the deviance values. For each production outcome, the mixed model contained the fixed effects of fatty acid variance, parity, their interaction, mean fatty acid proportion, and the random effect of month-year of calving.

Results

Overall, mean (SD) lactation number was 2.4 (1.4), representing cows from 1 to 9 lactations.

Mean milk production was 47.0 kg (9.2 kg), 50.4 kg (9.3 kg), and 46.4 kg (8.7 kg) at FT, PT, and MT, respectively (Table 1). Mean 305-day cumulative milk yield was 13,177 kg (2,041 kg). Proportion of de novo fatty acids was lowest at FT and greatest at MT, whereas proportion of preformed fatty acids was greatest at FT and lowest at MT (Table 1).

De novo fatty acids were significantly associated with milk and component yields across all periods. At FT, there was a significant interaction of de novo fatty acids and parity on test day and 305-day cumulative milk yields, in which the association

Table 1. Descriptive statistics of production performance and fatty acids (FA) at the selected lactation stages: first test (30 ± 2 DIM), peak test (68 ± 31 DIM), and mid test (100 ± 2 DIM).

	First Test		Peak Test		Mid Test	
Variable[1]	Mean	SD	Mean	SD	Mean	SD
De novo FA, % of fat	25.4	2.5	26.5	2.0	27.2	1.8
Mixed FA, % of fat	37.7	2.9	39.5	2.4	40.9	2.3
Preformed FA, % of fat	37.0	4.8	34.0	4.0	32.0	3.4
Test day yield, kg	47.0	9.2	50.4	9.3	46.4	8.7
Test day fat yield, kg	1.9	0.4	2.0	0.4	1.8	0.3
Test day ECM, kg	50.4	9.2	53.1	8.5	49.4	8.0

[1] Energy Corrected Milk = Yield × 0.3237 + Fat × 12.95 + Protein × 7.65.

was negative for both parties but more negative for primiparous animals ($P \leq 0.01$; Table 2). Test day and cumulative milk yields were negatively associated with de novo fatty acids at PT and were negatively associated and tended to be negatively associated with de novo fatty acids at MT, respectively ($P \leq 0.06$). Association of de novo fatty acids with component yields changed based on the DIM period. Milk fat yield and de novo fatty acids were negatively associated at FT ($P \leq 0.01$), not associated at PT ($P = 0.68$), and positively associated at MT ($P \leq 0.01$). Energy corrected milk yield followed similar patterns, although there was a significant interaction of de novo fatty acids and parity at FT, wherein ECM was negatively associated for both parities but more negative for primiparous animals. There was a significant interaction of de novo fatty acids and parity on milk protein yield at FT, in which multiparous animals had a positive association and primiparous had a negative association with yields of milk protein ($P \leq 0.01$). Later in lactation at MT, there was a positive association for all parities with de novo fatty acids and milk protein yield.

Preformed fatty acids were also significantly associated with milk and component yields across all periods. Generally, the opposite association was detected for preformed fatty acids compared with de novo fatty acids. Test day and 305-day cumulative milk yields were positively associated with preformed fatty acids across all three periods ($P \leq 0.01$). Yields of ECM were positively associated with preformed fatty acids at FT and PT but tended to be negatively associated at MT ($P \leq 0.07$). Milk fat yields were positively associated with preformed fatty acids at FT ($P \leq 0.01$) and negatively associated with preformed fatty acids at MT ($P \leq 0.01$), but an interaction between preformed fatty acids and parity was detected at PT ($P = 0.03$). The interaction effect indicates a positive association for multiparous cows and a negative association for primiparous cows at MT for fat yield. Different associations by parity were also observed for milk protein yield at FT, wherein yields of milk protein were negatively associated with multiparous cows and positively associated with primiparous cows for preformed fatty acids ($P = 0.02$). At PT and MT, milk protein yield was not associated with preformed fatty acids ($P > 0.1$).

Table 2. Least square means and standard errors of production performance for de novo fatty acids (FA) at the selected lactation stages: first test (30 ± 2 DIM), peak test (68 ± 31 DIM), and mid test (100 ± 2 DIM).

	FA		Parity		Interaction		P-value		
Yield[1], kg	Mean	SE	Mean	SE	Mean	SE	FA	Parity	Int.
First Test									
Milk	-0.39	0.14	1.19	6.76	-0.68	0.28	0.01	0.86	0.01
Fat	-0.03	0.01	-0.54	0.03			<0.01	<0.01	
Protein	0.009	0.004	0.23	0.18	-0.02	0.01	0.02	0.22	<0.01
ECM	-0.31	0.15	2.71	7.05	-0.71	0.29	0.04	0.70	0.01
305-day	-113	45	2682	2024	-234	82	0.01	0.19	<0.01
Peak Test									
Milk	-0.53	0.14	-14.72	0.62			<0.01	<0.01	
Fat	-0.002	0.01	-0.36	0.03			0.68	<0.01	
Protein	0.004	0.004	-0.33	0.02			0.27	<0.01	
ECM	-0.16	0.14	-11.92	0.67			0.24	<0.01	
305-day	-151	46	-3167	222			<0.01	<0.01	
Mid Test									
Milk	-0.39	0.18	-11.67	0.65			0.03	<0.01	
Fat	0.04	0.01	-0.30	0.02			<0.01	<0.01	
Protein	0.012	0.005	-0.28	0.02			0.01	<0.01	
ECM	0.44	0.16	-9.90	0.58			0.01	<0.01	
305-day	-128	67	-2817	234			0.06	<0.01	

[1] Energy Corrected Milk = Yield × 0.3237 + Fat × 12.95 + Protein × 7.65

Table 3. Least square means and standard errors of production performance for preformed fatty acids (FA) at the selected lactation stages: first test (30 ± 2 DIM), peak test (68 ± 31 DIM), and mid test (100 2 DIM).

	FA		Parity		Interaction		P-value		
Yield[1], kg	Mean	SE	Mean	SE	Mean	SE	FA	Parity	Int.
First Test									
Milk	0.36	0.06	-15.43	0.60			<0.01	<0.01	
Fat	0.01	0.003	-0.52	0.03			<0.01	<0.01	
Protein	-0.002	0.002	-0.73	0.15	0.01	0.004	0.22	<0.01	0.02
ECM	0.26	0.06	-14.62	0.63			<0.01	<0.01	
305-day	81	21	-7024	1622	104	42	<0.01	<0.01	0.01
Peak Test									
Milk	0.40	0.07	-14.83	0.61			<0.01	<0.01	
Fat	0.01	0.003	0.15	0.23	-0.01	0.01	0.06	0.52	0.03
Protein	0.000	0.002	-0.34	0.02			0.81	<0.01	
ECM	0.18	0.07	-12.14	0.66			0.01	<0.01	
305-day	104	23	-3186	215			<0.01	<0.01	
Mid Test									
Milk	0.37	0.09	-12.07	0.64			<0.01	<0.01	
Fat	-0.02	0.003	-0.30	0.02			<0.01	<0.01	
Protein	-0.003	0.002	-0.29	0.02			0.22	<0.01	
ECM	-0.16	0.08	-10.07	0.59			0.07	<0.01	
305-day	105	35	-2881	227			<0.01	<0.01	

[1] Energy Corrected Milk = Yield × 0.3237 + Fat × 12.95 + Protein × 7.65

Table 4. Association of within cow variance in de novo and preformed fatty acids with 28-day and 305-day cumulative milk yields.

Cumulative yield, kg	Estimate	SE	P-value
De novo			
28-day	81.1	54.0	0.14
305-day	1733.4	604.7	<0.01
Preformed			
28-day	-4.0	51.8	0.94
305-day	59.2	613.3	0.92

Conclusions

The proportion of de novo and preformed fatty acid groups exhibited considerable variation amongst cows both within week of lactation but also across lactation (Figure 1). Proportion of de novo fatty acids reached a maximum mean (SD) of 27.2% (1.8%) at 14 weeks of lactation and had a minimum mean of 23.8% (3.0%) at 2 weeks of lactation. Proportion of preformed fatty acids had the opposite relationship, wherein the minimum mean was reached at 16 weeks of lactation (31.5 ± 3.5%) and the maximum mean was achieved at 4 weeks of lactation (38.4 ± 5.2%). Individual cow variation in fatty acids across lactation was high. Variance in individual cow de novo proportion ranged from -0.66 to 0.85 and from -0.24 to 1.43 for preformed fatty acids. Individual cow lactational variance in de novo fatty acids were positively associated with 305-day cumulative milk yields ($P < 0.01$; Table 4). However, individual cow lactational variance in preformed fatty acids was not associated with either 28-day or 305-day cumulative milk yields ($P > 0.1$).

Milk fatty acids groups, de novo and preformed, are strongly associated with milk production outcomes at different stages of lactation. This suggests that these milk fatty

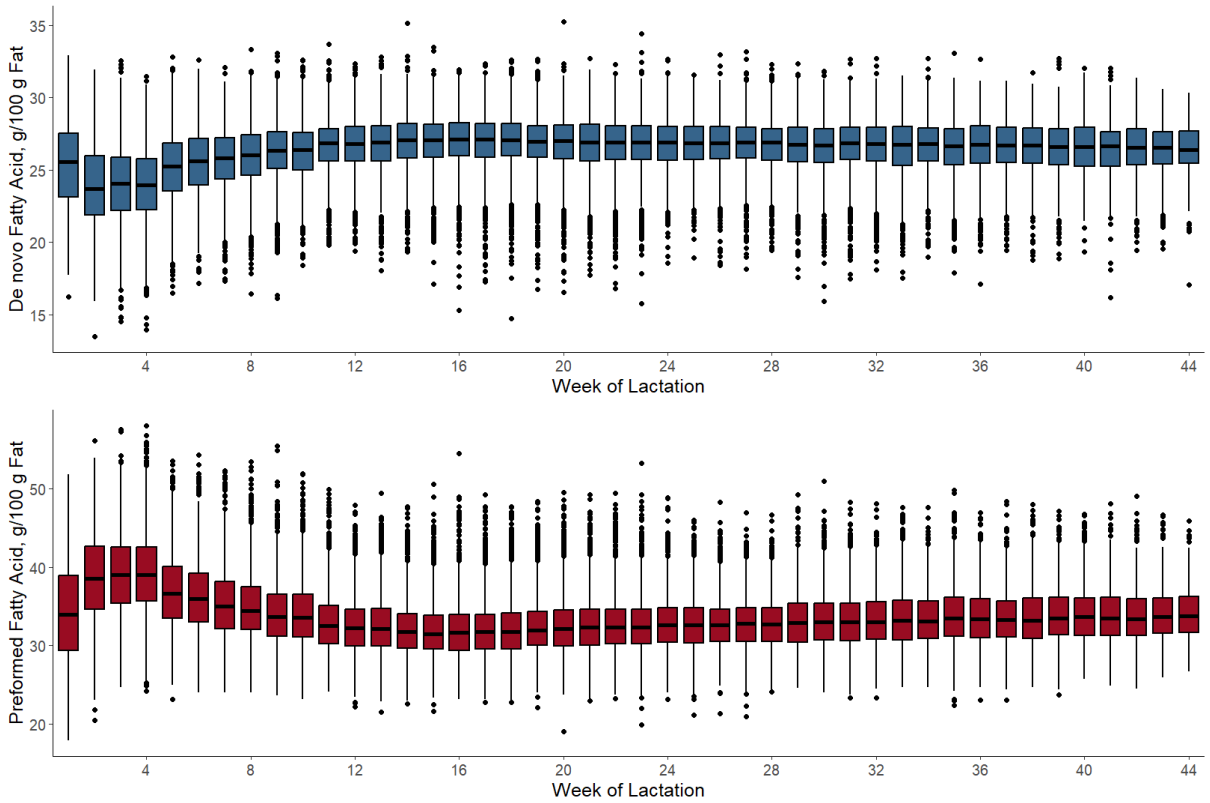

Figure 1. Box plots of de novo (top panel) and preformed (bottom panel) fatty acids of individual cows by week of lactation.

acids may be a useful management tool to aid in making pen grouping decisions, cow selection and breeding decisions, and informing dietary adjustments. Changes in the direction of the association between milk fatty acids and milk components at different lactation stages, as well as interactions with parity, underscores the importance of considering these factors when making management decisions based on a single milk test. A positive association of individual cow lactational variance in de novo fatty acids with 305-day cumulative milk yields is an interesting finding and warrants further investigation to understand the biology behind this association. Combined, the results herein indicate that routine milk testing across lactation may allow for tailored management decisions at the individual cow level using these fatty acid groups.

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