Herd Management Milk Analysis: Jersey versus Holstein and Between Lab Agreement of Results

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Introduction

In 2018 (Barbano et al., 2018), we provided a summary of how de novo, mixed, and preformed milk fatty acid concentrations in milk measured by mid-infrared (**MIR**) changed in relation to bulk tank fat and protein test for Holstein dairy herds. The analytical aspects of reference milk fatty acid (**FA**) analysis and model development and validation were reported by Wojciechowski and Barbano (2016) and Woolpert et al. (2016). The form of the FA data from the MIR (Barbano et al., 2014, 2017) was structured to provide information on the relative proportions of de novo (C4 to C15), mixed origin (C16:0, C16:1, C17:0), and preformed (C18:0 and longer) FA in milk, the mean FA chain length (carbon number) and degree of unsaturation (double bonds/fatty acid). Since that time, we have continued to collect data on milk FA variation in bulk tank milk for Jersey herds.

Woolpert et al. (2016, 2017) have reported the results of two studies to determine feeding and farm management factors influencing milk FA composition and their relationship to bulk tank milk fat and protein test and production per cow per day of fat and protein. In the first study (Woolpert et al., 2016) 44 commercial dairies were identified as either predominantly Holstein or Jersey in northern Vermont and New York. The yields of milk fat, true protein, and de novo FA per cow per day were higher for high de novo (**HDN**) versus low de novo (**LDN**) farms. Woolpert et al. (2016, 2017) estimated the impact of differences in de novo fatty acid concentration in milk among farms on bulk tank fat and protein, and estimated the impact of those differences on farm income per 100 cows per year. Higher milk de novo fatty concentration drove higher milk fat, milk protein, and grew revenues from milk. A study of Jersey herds from around the US was conducted during the past year to provide a comparison of milk fatty acid data mean data for a 16 month period for Holstein herds from the Northeast versus Jersey herds studied monthly for a 12 month period from different regions of the US. The relationship of milk fatty acid composition to bulk tank milk fat and protein test for both breeds of cattle is reported in this paper.

These relationships of milk fatty acid composition to bulk tank milk fat and true protein concentration are the basis of use of milk fatty acid composition in combination with herd management information to aid in making decisions to adjust dairy cattle ration composition or management to improve the production of milk fat, protein, and milk volume per cow per day. It has been shown that seasonal variation of fat and protein concentration in bulk tank milks in the northeastern US is related to seasonal varation in de novo fatty acid concentration and production in grams per cow per day (Barbano et al., 2017).

Our objective in the current work was to measure and compare the relationships of milk fatty acid composition and bulk tank fat and true protein test to milk fatty acid composition for Holstein versus Jersey farms. Based on data from our previous studies the following graphs (Figures 1,3,5,7,9 11, 13) for Holstein farms were developed to help farms understand the relationships between bulk tank milk FA composition and bulk tank milk fat and protein test. In the current paper, new data on bulk tank fat and protein tests and their relationship to milk fatty acid composition for bulk tank milk on Jersey farms (Figures 2, 4, 6, 8, 10, 12) are presented.

De Novo Fatty Acids and Milk Fat

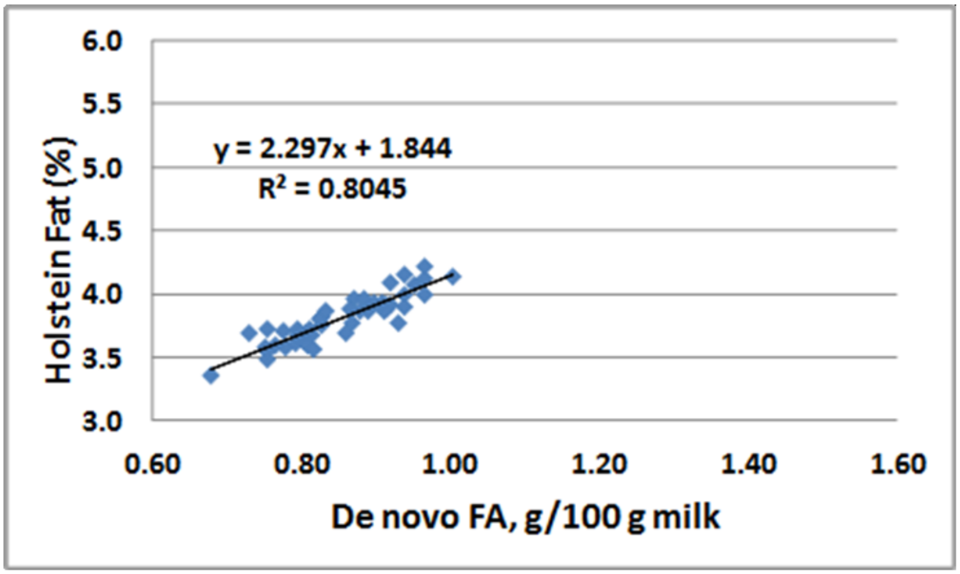

Figure 1. Relationship of bulk tank milk fat test to concentration (g/100 g milk) of de novo FA in Holstein herd bulk tank milk.

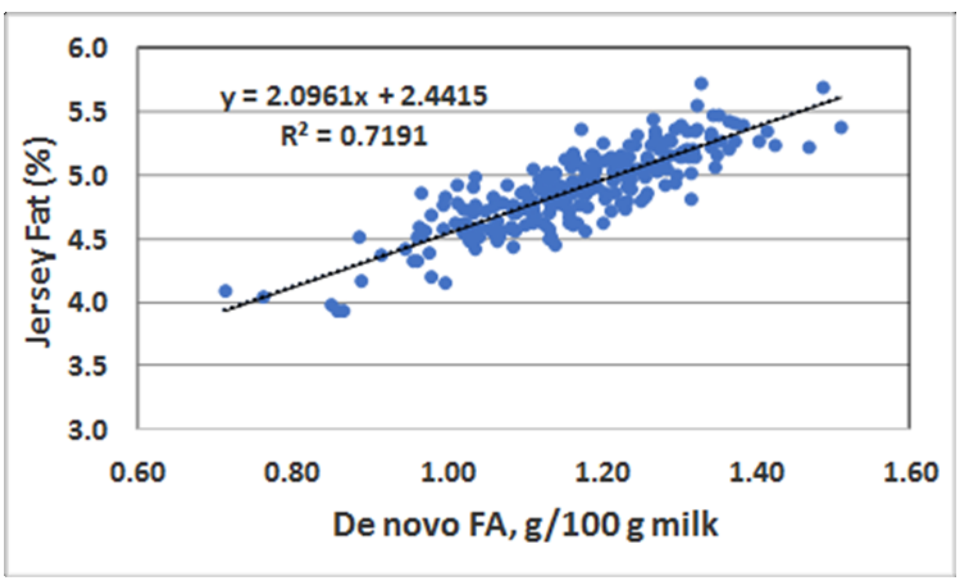

Figure 2. Relationship of bulk tank milk fat test to concentration (g/100 g milk) of de novo FA in Jersey herd bulk tank milk.

Bulk tank milk fat concentration increases significantly ($P < 0.05$) with increasing de novo fatty acid concentration in milk for both Holstein and Jersey cattle with the slopes of these relationships being very similar.

Mixed Origin Fatty Acids and Milk Fat

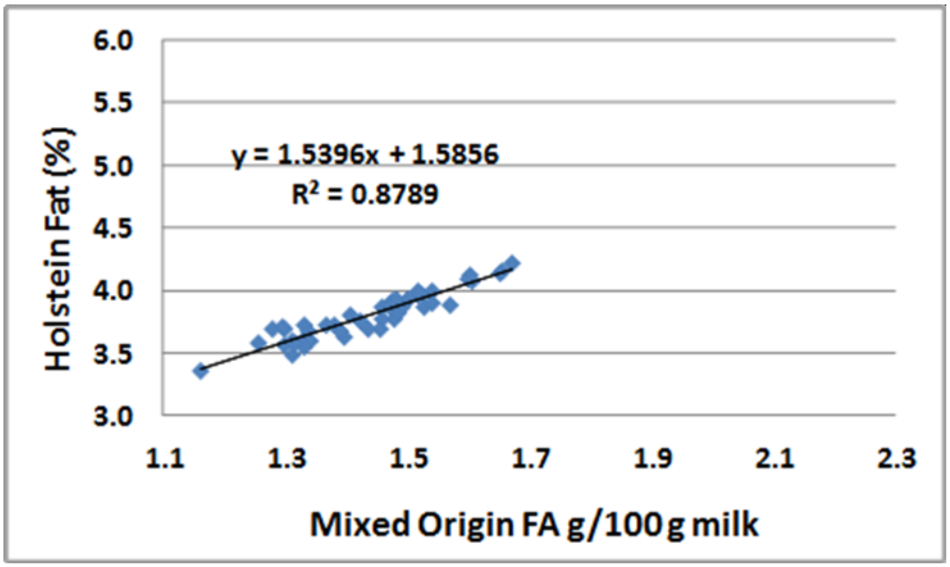

Figure 3. Relationship of bulk tank milk fat test to concentration (g/100 g milk) of mixed origin FA in Holstein bulk tank milk.

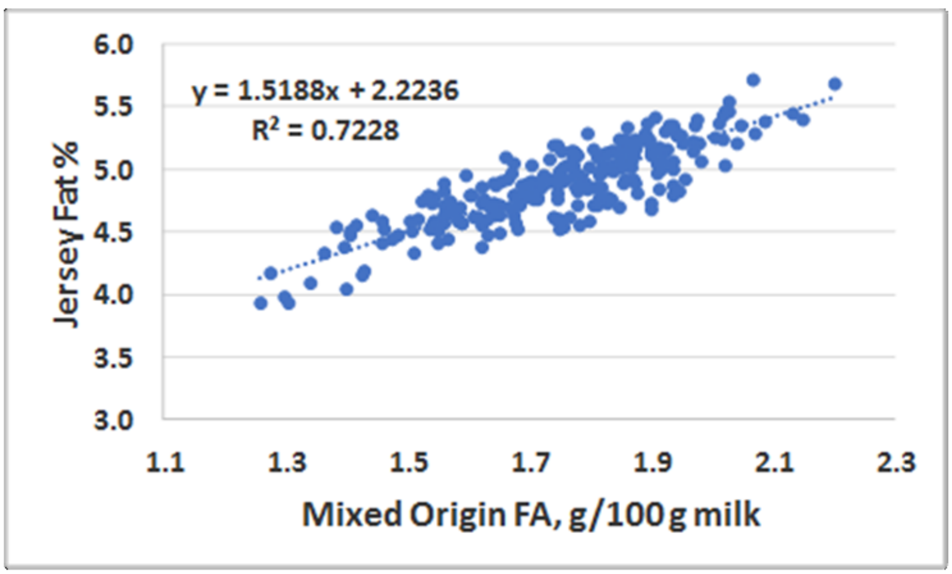

Figure 4. Relationship of bulk tank milk fat test to concentration (g/100 g milk) of mixed origin FA in Jersey bulk tank milk.

Similar to what was observed for de novo fatty acids (Fgiures 1 and 2) bulk tank milk fat concentration increases significantly (P < 0.05) with increasing mixed origin fatty acid concentration in milk for both Holstein and Jersey cattle (Figures 3 and 4) with the slopes of these relationships being very similar.

De Novo plus Mixed Origin Fatty Acids and Milk Fat

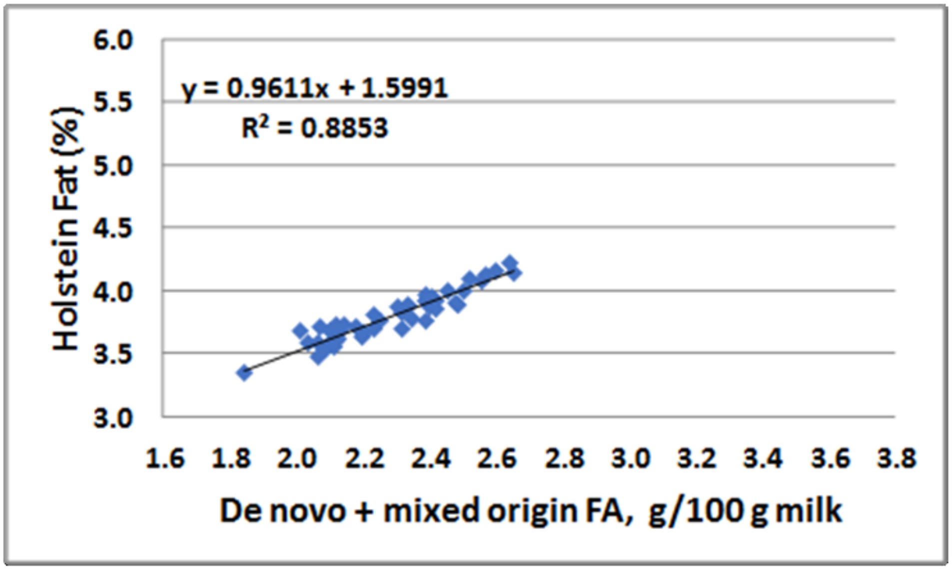

Figure 5. Relationship of bulk tank milk fat test to concentration (g/100 g milk) of de novo plus mixed origin FA in Holstein bulk tank milk.

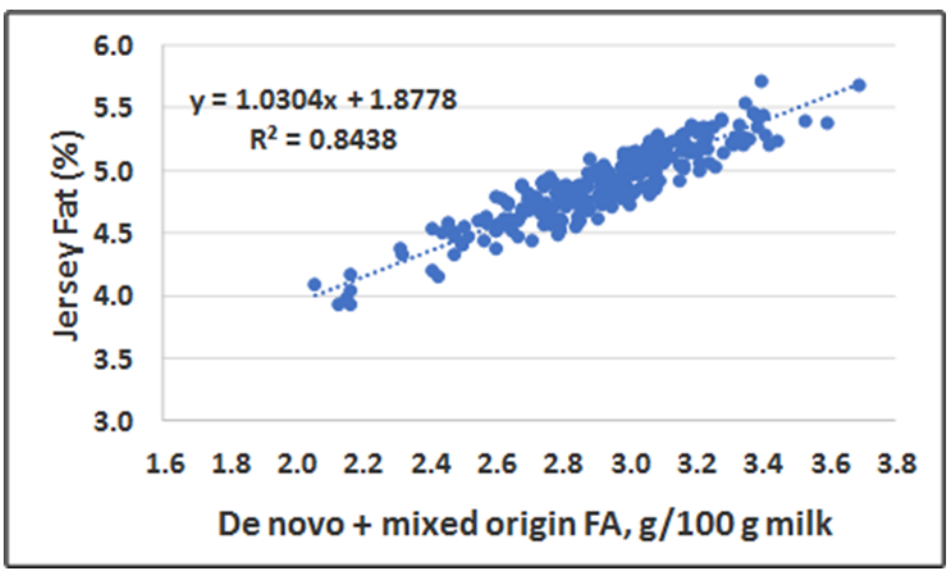

Figure 6. Relationship of bulk tank milk fat test to concentration (g/100 g milk) of de novo plus mixed origin FA in Jersey bulk tank milk.

Bulk tank milk fat concentration increases significantly (P < 0.05) with increasing de novo plus mixed origin fatty acid concentration in milk for both Holstein and Jersey cattle with the slopes of these relationships being very similar. The R^2 for these correlations are high and consistent among the two breeds of cattle. On average a herd that does not have a seasonal calving pattern the average days in milk for the herd would be in the range of 150 to 200 days in milk. Therefore, on average the cows have a net positive energy balance and the bulk tank milk composition will be more strongly influenced by milk from cows in positive energy balance. With increasing days since calving the proportion of the palmitic acid (C16:0) in milk shifts palmitic acid originating from adipose tissue to palmitic acid be produced by the de novo synthesis pathway. Transfer of palmitic acid from by pass fat feeding when cow are in positive may impact this relationship and the ratio of mixed to de novo milk fatty acids.

Preformed Fatty Acids and Milk Fat

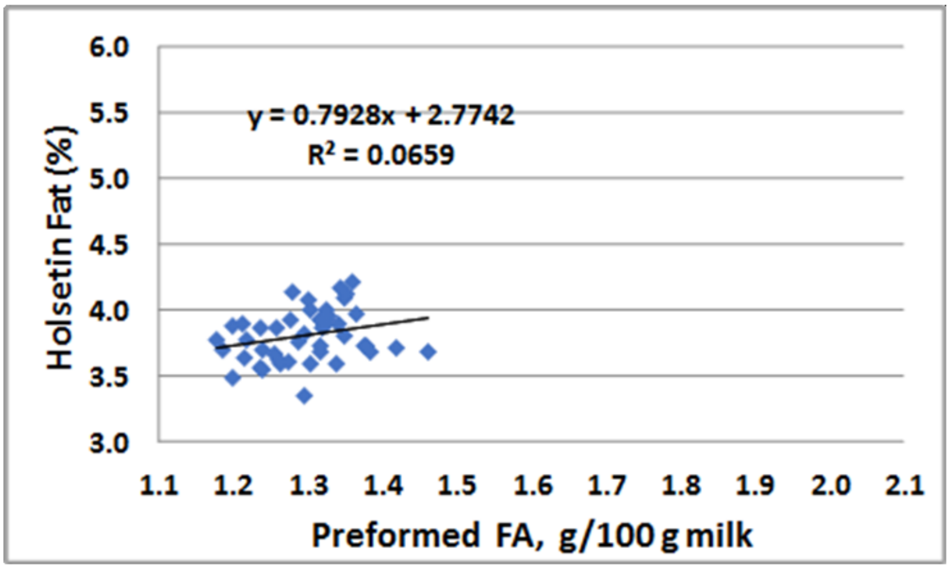

Figure 7. Relationship of Holstein bulk tank milk fat test to concentration (g/100 g milk) of preformed FA in bulk tank milk.

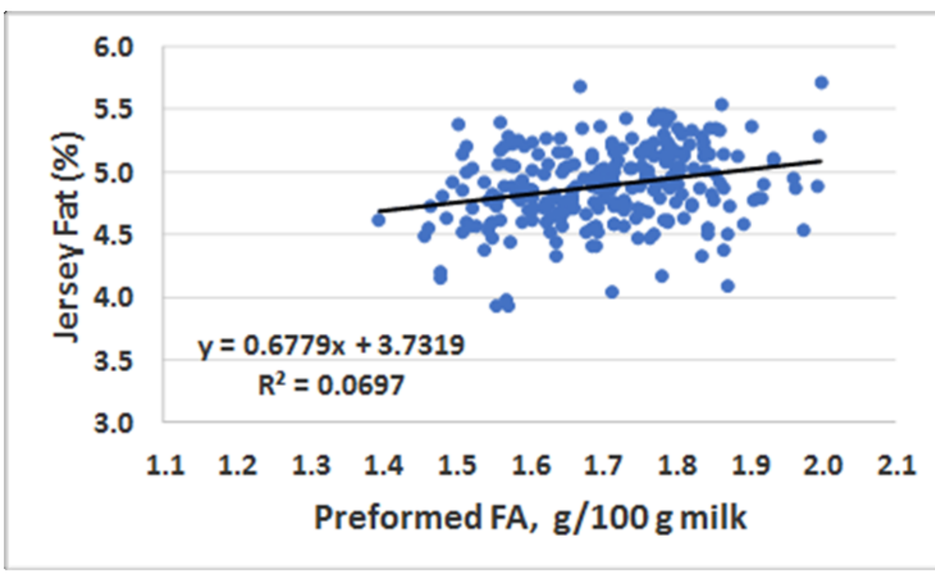

Figure 8. Relationship of Jersey bulk tank milk fat test to concentration (g/100 g milk) of preformed FA in bulk tank milk.

No increase in bulk tank milk fat concentration with increasing preformed fatty acid concentration in milk for both Holstein and Jersey cattle was detected ($P > 0.05$) with the slopes of these relationships not being significantly different from zero.

Double Bonds per Fatty Acid (Milk fat depression index) and Milk Fat

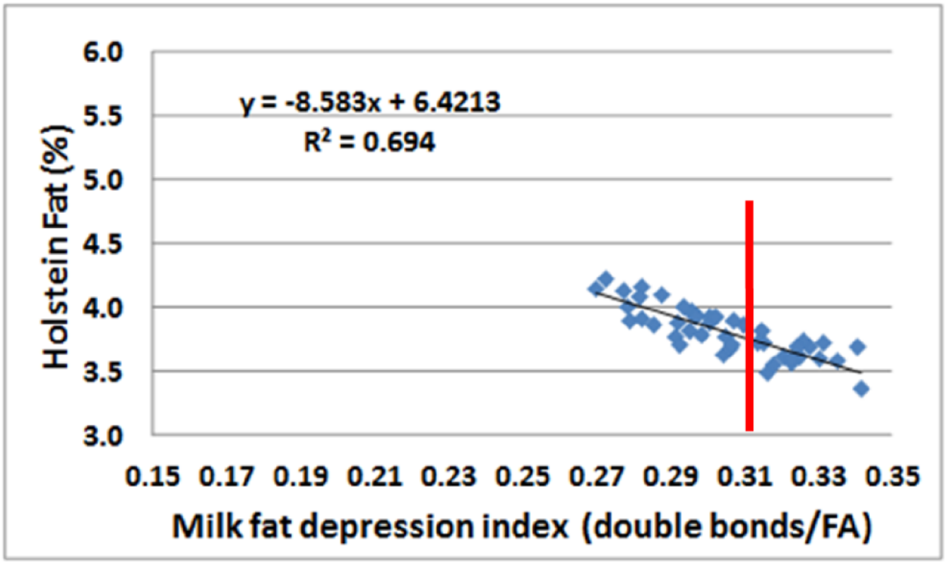

Figure 9. Milk fat depression index for <u>Holstein</u> bulk tank milk fatty acid unsaturation with bulk tank milk fat test. As double bonds per fatty acid increases the bulk tank milk fat test decreases. When double bond per fatty acid values are higher than the vertical line, there is a higher probability of unsaturated fat being too high or being released too fast in the ration.

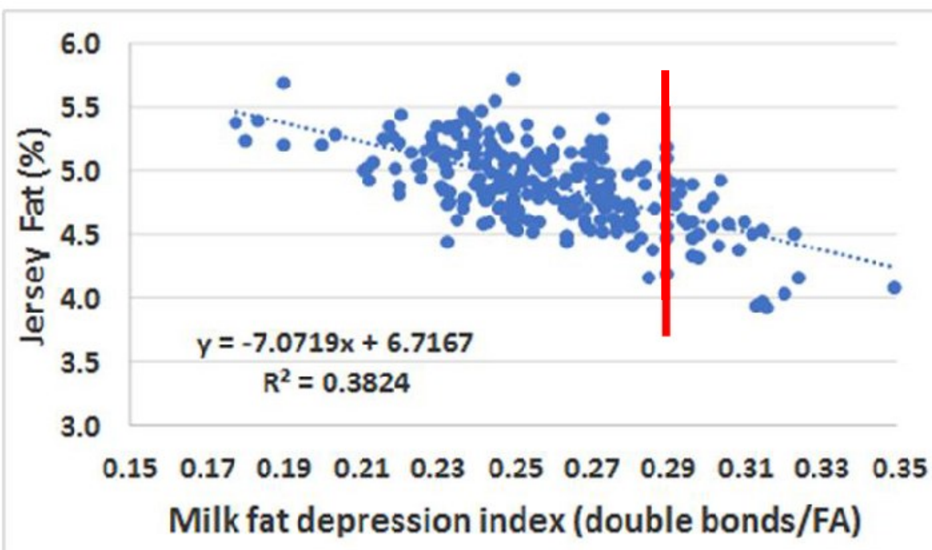

Figure 10. Milk fat depression index <u>Jersey</u> bulk tank milk fatty acid unsaturation with bulk tank milk fat test. When double bond per fatty acid values are higher than the vertical line, there is a higher probability of unsaturated fat being too high or being released too fast in the ration.

The PLS model for direct measurement of double bonds per fatty acid in milk fat was reported by Wojciechowski et (2016). As double bonds per fatty acid increases, the bulk tank milk fat test decreases. We have documented this in both Holstein and Jersey milks. In the Holstein milks represented in Figure 9, we have measured the level of C18:1 trans 10 fatty acid (with gas lIquid chromatorgraphy) and in Holstein milks with a double bond per fatty acid higher than 0.31, the level of C18:1 trans 10 fatty acid is elevated indicating trans fatty acid induced milk fat depression. This is consistent with the report of Harvatine and Bauman (2011) that elevated levels of C18:1 trans 10, cis 12 CLA was related to milk fat depression. Similar results based on GLC analysis is found for Jersey cows, however the mean double bonds per fatty acid is lower for Jersey than for Holstein milk. Double bonds per fatty acid is an index and a high value for double bonds per fatty acid is an indicator of trans fatty acid induced milk fat depression. This is a valuble piece of farm management information when trying to interpret why overall milk fat percentage is low. If fat percent is low and the double bonds per fatty acid is low, then it is likely that the cause of the low fat is not classical trans fatty acid induced milk fat depression. Other causes of low milk fat may be low dry mater intake or other animal health issues that have caused immune system activation resulting in high demand for glucose to support the immune system response.

De Novo Fatty Acids and Milk Protein

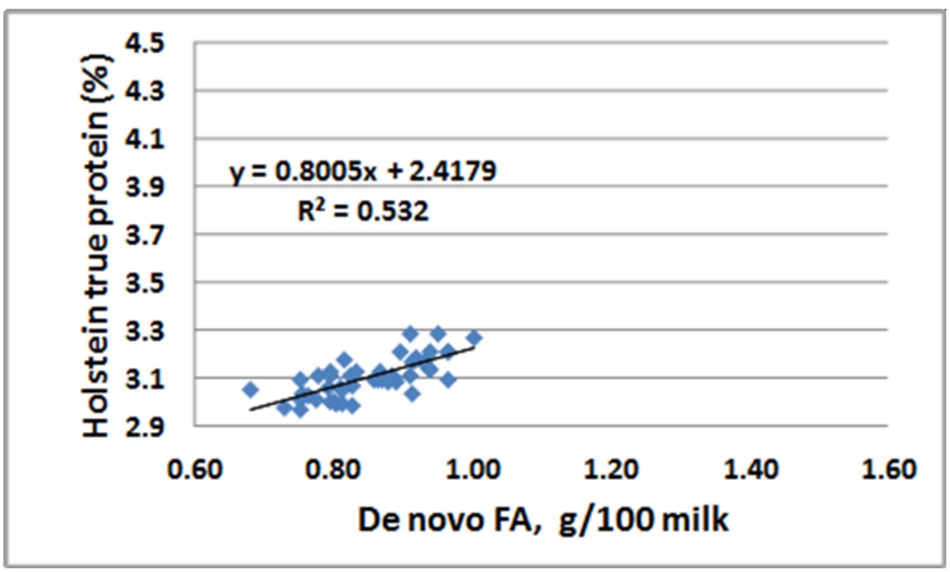

Figure 11. Relationship of <u>Holstein</u> bulk tank milk true protein concentration with change in de novo milk fatty acid concentration.

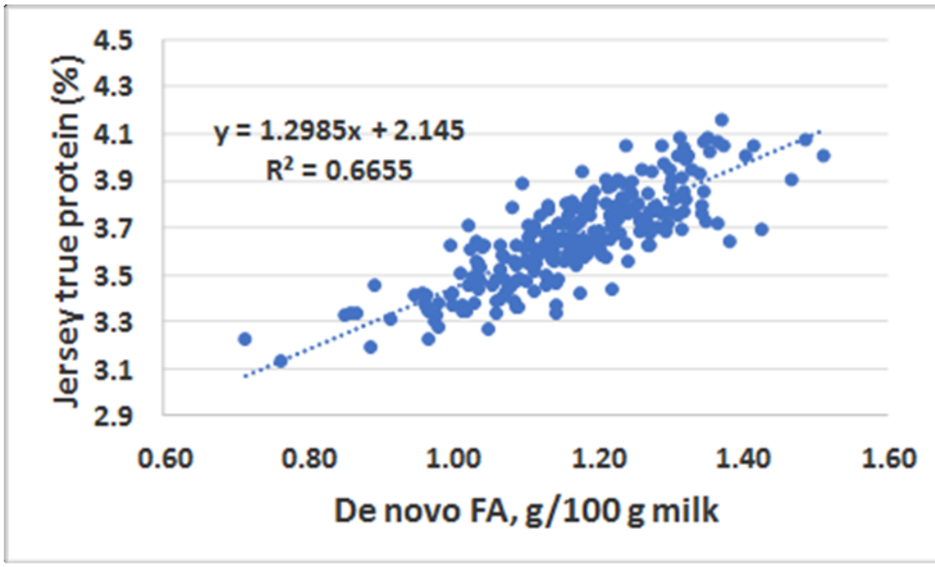

Figure 12. Relationship of Jersey bulk tank milk true protein concentration with change in de novo milk fatty acid concentration.

Milk true protein concentration increases with increasing milk de novo fatty acid concentration for both Holstein (Figure 11) and Jersey (Figure 12) bulk tank milks. Jersey have a larger increase in milk protein production per 100 grams of milk in response to increased denovo fatty acid production than Holsteins. Woolpert reported (2016, 2017) that dairy herds that had higher milk de novo fatty concentration produced more grams of protein per cow per day than herds that had low de novo fatty acid concentration in milk.

Preformed Fatty Acids and Milk Protein

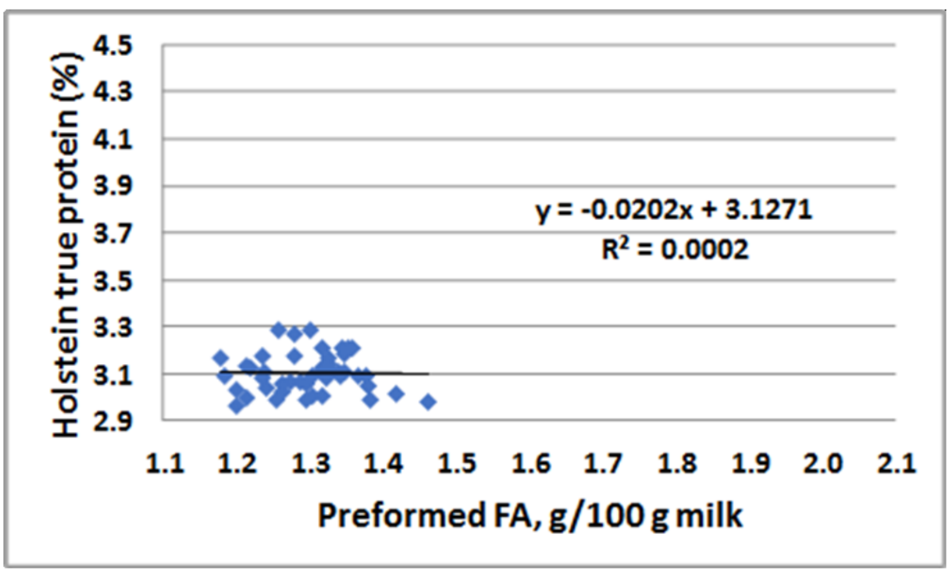

Figure 13. Relationship of Holstein bulk tank milk true protein concentration with change in preformed milk fatty acid concentration.

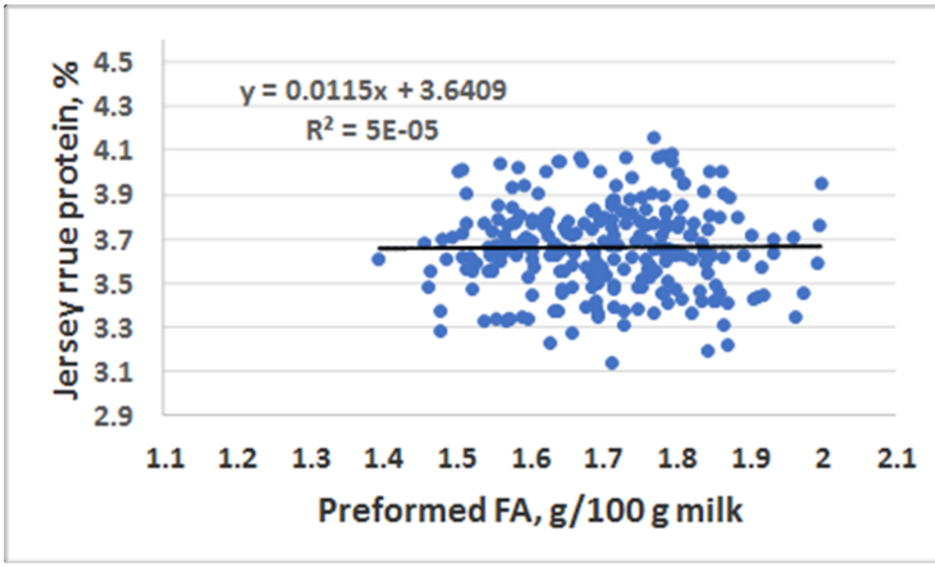

Figure 14. Relationship of Jersey bulk tank milk true protein concentration with change in preformed milk fatty acid concentration. As de novo milk fatty acid concentration increases milk protein increases.

As preformed milk fatty acid concentration increased no change in milk protein concentration was detected ($P > 0.05$) in either Holstein or Jersey bulk tank milk.

Between Lab Agreement Among Laboratories: Milk Fatty Acid Testing

Milk fatty acid prediction models need to be calibrated with reference milks that have reference values established by gas chromatography analysis (like calibration done for total fat, protein and lactose). The milk fatty acid GLC method calculation of results of GLC results for individual fatty acids, fatty acid chain length, and fatty acid double bonds per fatty acid used for this research was described by Wojceichowski et al. (2016) and Kaylegian et al. (2009 a,b). For this work only major fatty acids defined for the MIR application are included in the calculations and GLC data are normalized to 100% to ensure better lab-to-lab consistency of results of reference chemistry. This standardized approach will ensure better agreement among laboratories in both reference chemistry and MIR results.

Calibration samples for milk fatty acid analysis by MIR are produced at Cornell once every 4 weeks. The production of the calibration samples was described by Kaylegian et al., (2006a) and their performance for calibration of MIR milk analyzers for measuring milk fat, protein, lactose and solids was reported and compared to the use of individual farm milks (Kaylegian et al., 2006b). This same set of samples that is used across the US for calibration of MIR instruments for milk components is being used to calibrate MIR milk analyzers for milk fatty acid analysis with reference values expressed in grams per 100 gram of milk. The calibration set has a wide range of concentration of individual fatty acids and groups of fatty acids (e.g., de novo, mixed origin, and preformed fatty acids).

To validate the performance of a group of 9 MIR milk analyzers (Delta Instruments models FTA, combi 300, and combi 600 instruments), that were set up using the milk fatty acid PLS models for direct measurement of fatty acid chain length and double bonds per fatty acid (Wojceichowski et al., 2016) and the PLS models for direct measurement of the groups of fatty acids defined as de novo, mixed origin, and preformed fatty acids reported by Woolpert et al. (2016). These instruments were calibrated (i.e., adjustment of secondary slope and intercept) for de novo, mixed orgin and performed fatty acid once every 4 weeks using the modified milk calibration samples (14 sample set) produced at Cornell University. The fatty acid chain length and double bonds per fatty acid models were calibrated periodically with a set of 8 individual producer milk samples with defined reference values. The instruments were calibrated approximately 2 weeks prior to the testing of the set of unknown validation samples. The validation samples were 8 individual farm milks. There were 2 farm milks from each of 4 different geographic regions of the US. This set of validation milks was tested by gas chromatography to establish reference values and was tested on each of 9 different MIR milk analyzers in different regions of the US.

A typical example of the reference chemistry for set of modified milk calibration samples for milk fatty acids is shown below in Table 1.

Table 1. Modified milk calibration sample reference chemistry.

Sample	total grams de novo fatty acid (g/100g milk)	total grams mixed origin fatty acid (g/100g milk)	total grams preformed fatty acid (g/100g milk)	total grams fat (g/100g milk)
1	0.0471	0.0846	0.0731	0.2167
2	0.1400	0.2514	0.2174	0.6438
3	0.2347	0.4215	0.3644	1.0793
4	0.3291	0.5912	0.5111	1.5139
5	0.4231	0.7600	0.6570	1.9461
6	0.5150	0.9250	0.7997	2.3688
7	0.6091	1.0941	0.9459	2.8017
8	0.7028	1.2624	1.0914	3.2326
9	0.7968	1.4311	1.2373	3.6648
10	0.8896	1.5979	1.3814	4.0918
11	0.9844	1.7681	1.5287	4.5278
12	1.0767	1.9340	1.6721	4.9526
13	1.1722	2.1055	1.8204	5.3918
14	1.2629	2.2685	1.9612	5.8090
Mean	0.6560	1.1782	1.0187	3.0172
min	0.0471	0.0846	0.0731	0.2167
max	1.2629	2.2685	1.9612	5.8090
Range	1.2158	2.1839	1.8881	5.5924

The results of the MIR milk analysis of the validation samples is given in Tables 2 through 6 below.

Table 2. Reference values and individual laboratory predictions for de novo fatty acid concentration (g/100 g milk) in 8 individual farms milk validation samples and calculated mean difference (MD) and standard deviation of the difference (SDD) from the reference values fore each of 9 different laboratories running Delta FTA or Delta Combi MIR milk analyzers.

	denovo		Lab	Lab	Lab	Lab	Lab	Lab	Lab	Lab	lab
Sample	Reference		1	2	3	4	5	6	7	8	9
1	0.8991		0.860	0.862	0.874	0.860	0.870	0.894	0.920	0.890	0.890
2	0.8484		0.820	0.810	0.838	0.820	0.822	0.828	0.840	0.820	0.830
3	0.7209		0.720	0.732	0.743	0.730	0.715	0.748	0.750	0.720	0.720
4	0.8179		0.810	0.811	0.819	0.800	0.789	0.804	0.840	0.800	0.830
5	0.7540		0.720	0.729	0.754	0.750	0.731	0.740	0.740	0.730	0.740
6	0.9635		0.930	0.937	0.964	0.940	0.933	0.953	0.950	0.930	0.950
7	0.7910		0.810	0.798	0.803	0.820	0.796	0.804	0.840	0.810	0.810
8	1.3033		1.220	1.224	1.252	1.240	1.234	1.220	1.240	1.230	1.250
	0.8873	Mean	0.861	0.863	0.881	0.870	0.861	0.874	0.890	0.866	0.878
		MD	-0.026	-0.024	-0.006	-0.017	-0.026	-0.013	0.003	-0.021	-0.010
		SDD	0.031	0.029	0.023	0.029	0.022	0.032	0.035	0.027	0.022

The agreement of mean values among MIR instruments for de novo fatty acids was excellent and they were in good agreement with the GLC reference chemistry for these samples.

Table 3. Reference values and individual laboratory predictions for mixed origin fatty acid concentration (g/100 g milk) in 8 individual farms milk validation samples and calculated mean difference (MD) and standard deviation of the difference (SDD) from the reference values fore each of 9 different laboratories running Delta FTA or Delta Combi MIR milk analyzers.

	Mixed		Lab	Lab	Lab	Lab	Lab	Lab	Lab	Lab	lab
	Reference		1	2	3	4	5	6	7	8	9
1	1.3295		1.480	1.445	1.438	1.420	1.419	1.471	1.480	1.490	1.460
2	1.1070		1.220	1.170	1.162	1.180	1.163	1.168	1.170	1.220	1.200
3	0.9481		1.050	1.042	1.041	1.010	0.996	1.035	1.030	1.060	1.040
4	1.1063		1.240	1.232	1.208	1.210	1.158	1.186	1.260	1.260	1.230
5	1.0260		1.100	1.098	1.103	1.100	1.049	1.078	1.070	1.100	1.080
6	1.3599		1.490	1.455	1.472	1.440	1.414	1.482	1.440	1.450	1.460
7	1.3105		1.330	1.261	1.267	1.300	1.227	1.225	1.290	1.300	1.280
8	1.5220		1.660	1.625	1.648	1.640	1.580	1.630	1.650	1.680	1.620
Mean	1.2136		1.321	1.291	1.292	1.288	1.251	1.285	1.299	1.320	1.296
		MD	0.108	0.077	0.079	0.074	0.037	0.071	0.085	0.106	0.083
		SDD	0.043	0.055	0.054	0.039	0.052	0.070	0.059	0.057	0.051

Overall, the between lab agreement of MIR instruments for mixed orgin fatty acids was good but the mean estimate by the group of instruments was a bit higher (0.08 g/100 g of milk) than GLC reference chemistry on this group of validation samples.

Table 4. Reference values and individual laboratory predictions for preformed fatty acid concentration (g/100 g milk) in 8 individual farms milk validation samples and calculated mean difference (MD) and standard deviation of the difference (SDD) from the reference values for each of 9 different laboratories running Delta FTA or Delta Combi MIR milk analyzers.

	Preformed		Lab	Lab	Lab	Lab	Lab	Lab	Lab	Lab	lab
	Reference		1	2	3	4	5	6	7	8	9
1	1.4988		1.370	1.419	1.426	1.480	1.451	1.405	1.410	1.380	1.390
2	1.4982		1.390	1.479	1.492	1.450	1.468	1.484	1.470	1.400	1.440
3	1.5371		1.410	1.438	1.427	1.460	1.480	1.458	1.470	1.390	1.490
4	1.5798		1.440	1.471	1.544	1.510	1.561	1.563	1.430	1.400	1.490
5	1.4224		1.370	1.371	1.370	1.380	1.438	1.429	1.440	1.350	1.460
6	1.7128		1.560	1.635	1.606	1.690	1.677	1.622	1.660	1.620	1.660
7	1.3716		1.310	1.414	1.434	1.370	1.442	1.477	1.410	1.340	1.400
8	1.7819		1.690	1.739	1.695	1.750	1.784	1.774	1.730	1.650	1.760
Mean	1.5503		1.443	1.496	1.499	1.511	1.538	1.526	1.503	1.441	1.511
		MD	-0.108	-0.055	-0.051	-0.039	-0.013	-0.024	-0.048	-0.109	-0.039
		SDD	0.036	0.049	0.058	0.026	0.041	0.066	0.059	0.046	0.052

Overall, the between lab agreement of MIR instruments for preformed fatty acids was good but the mean estimate by the group of instruments was a bit lower (0.054 g/100 g of milk) than GLC reference chemistry on this group of validation samples. It has been our experience on prediction of mixed and preformed milk fatty acids with the first generation of PLS prediction models on the Delta instruments, that when the models for mixed origin and preformed milk fatty acids do not agree with GLC reference chemistry they are off in opposite directions. A second generations of fatty acid PLS models is under development.

Table 5. Reference values and individual laboratory predictions for mean fatty acid chain length (carbon number) in 8 individual farms milk validation samples and calculated mean difference (MD) and standard deviation of the difference (SDD) from the reference values for each of 9 different laboratories running Delta FTA or Delta Combi MIR milk analyzers.

	CL		Lab	Lab	Lab	Lab	Lab	Lab	Lab	Lab	Lab
Sample	Reference		1	2	3	4	5	6	7	8	9
1	14.7434		14.63	14.76	14.80	14.72	14.65	14.65	14.67	14.76	14.76
2	14.7429		14.64	14.78	14.79	14.69	14.61	14.69	14.71	14.77	14.78
3	14.8803		14.75	14.85	14.91	14.83	14.76	14.73	14.82	14.88	14.88
4	14.7634		14.64	14.72	14.76	14.68	14.64	14.65	14.64	14.77	14.73
5	14.7897		14.67	14.75	14.78	14.71	14.66	14.67	14.73	14.76	14.78
6	14.8062		14.61	14.74	14.77	14.69	14.63	14.61	14.70	14.77	14.77
7	14.7861		14.67	14.79	14.83	14.73	14.68	14.69	14.73	14.76	14.82
8	14.4498		14.32	14.38	14.46	14.37	14.25	14.32	14.32	14.43	14.47
Mean	14.7452		14.616	14.721	14.763	14.678	14.610	14.626	14.665	14.738	14.749
		MD	-0.129	-0.024	0.017	-0.068	-0.135	-0.119	-0.080	-0.008	0.004
		SDD	0.029	0.039	0.032	0.028	0.037	0.043	0.035	0.023	0.028

The 9 instruments in the validation study had not had a calibration adjustment for milk fatty acid chain length (Table 5) or double bonds per fatty acid (Table 6) in 8 months. It has been our experience that the calibration PLS models for prediction of structural parameters such as fatty acid chain length and double bonds per fatty acid are much more stable across time (versus concentration parameters). The agreement of among instruments for mean fatty acid chain length was good with the mean value about 0.06 carbons lower than the reference value for this set of validation samples.

Table 6. Reference values and individual laboratory predictions for mean fatty acid unsaturation (double bonds per fatty acid) in 8 individual farms milk validation samples and calculated mean difference (MD) and standard deviation of the difference (SDD) from the reference values for each of 9 different laboratories running Delta FTA or Delta Combi MIR milk analyzers.

	DB/FA		Lab	Lab	Lab	Lab	Lab	Lab	Lab	Lab	Lab
Sample	Reference		1	2	3	4	5	6	7	8	9
1	0.2651		0.260	0.275	0.289	0.270	0.281	0.277	0.260	0.290	0.270
2	0.2974		0.290	0.308	0.318	0.288	0.301	0.310	0.300	0.310	0.300
3	0.3405		0.320	0.329	0.344	0.326	0.334	0.328	0.330	0.340	0.340
4	0.2987		0.290	0.299	0.311	0.291	0.307	0.309	0.290	0.310	0.300
5	0.3237		0.310	0.316	0.325	0.305	0.319	0.321	0.310	0.320	0.320
6	0.3065		0.290	0.299	0.310	0.286	0.301	0.293	0.300	0.310	0.300
7	0.2841		0.280	0.302	0.311	0.282	0.302	0.306	0.290	0.300	0.300
8	0.2649		0.250	0.255	0.273	0.245	0.259	0.268	0.250	0.260	0.250
Mean	0.2976		0.286	0.298	0.310	0.287	0.301	0.302	0.291	0.305	0.298
		MD	-0.011	0.000	0.013	-0.011	0.003	0.004	-0.006	0.007	0.000
		SDD	0.006	0.011	0.010	0.009	0.010	0.013	0.007	0.010	0.009

The agreement of among instruments for mean fatty acid unsaturation was excellent both from instrument to instrument and in agreement with reference chemistry for this set of validation samples.

Take Away Messages

1) The relationships documented among milk fatty acid composition and bulk tank milk fat and true protein test in Jersey milk follows the same relationships that have been documented for Holstein dairy herds.

2) Jersey cows produce more milk fat and tend to have a higher relative concentration of de novo and mixed origin fatty acids than observed for Holstein cows.

3) Jersey cows seem to have a larger increase in milk true protein concentration than Holstein cows as milk de novo fatty concentration increases.

4) Jersey cows have a shorter mean milk fatty acid chain length and lower mean unsaturation than milk from Holstein cows. However, the relationship of decreasing fat and protein concentration in milk with increasing mean unsaturation is similar in both breeds and higher level of mean double bonds per fatty acid is an indicator of trans fatty acid induced milk fat depression.

5) Between lab agreement between the MIR compared in this study milk fatty acid analysis was very good for instruments that were calibrated every 4 weeks with reference. The between laboratory agreement was best for de novo fatty acid content (g/100 g milk) and milk fat depression index (double bonds per fatty acid).

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