What do Clues in Milk Composition Parameters Tell us About Herd Performance?

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A continuing effort in analyzing fat composition has opened a new frontier in evaluating herd performance (Woolpert, et al, 2016). For a long time, bulk tank milk has been analyzed for total fat content. Herd managers soon realized that feeding too much grain or putting cows on pasture would cause a depression in milk fat. These causes of milk fat depression were dogma until it was demonstrated that a potent unsaturated fat (C18:2 trans-10, cis-12) was responsible for many cases of milk fat depression (Bauman and Griinari, 2003).

In 1978, total protein was added to routine bulk tank milk analysis which was later refined to be true protein (January 2000) in most Federal Milk Marketing Orders, although California (not part of the Federal Orders) remains on the total protein system. Interest in milk protein increases in areas with multiple component pricing and when the price of protein is high (> $2.00/lb). About the same time, milk urea nitrogen (MUN) was added to routine milk analysis but has not been incorporated into a payment system. MUN is generally a measure of excess urea in the rumen. As a guideline, a MUN value of 5-6 has been shown to indicate a rumen that is short of available nitrogen while values above 15 usually indicate excessive nitrogen excretion.

The latest addition to milk component analysis is the identification of specific milk fatty acids (Woolpert, et. Al., 2016). Because milk fat is in the form of triglycerides, there is a glycerol backbone that constitutes approximately 5.5% of the total fat weight. Therefore, fatty acids represent approximately 94.5% of the milk fatty acid weight. There are two main sources of fatty acids. The first is de novo synthesis by the mammary cell which can fatty acids from 4 to 16 carbons long. This occurs as a result of elongating acetate and butyrate (produced in the rumen and transported to the mammary cell via the blood) into fatty acids. However, butyrate is the foundation for nearly all de novo synthesis. Preformed fatty acids, originating from either the diet or from adipose stores in the body, are transported into the mammary cell. Preformed fatty acids are 16 carbons and longer. These two sources overlap in the 16 carbon fatty acids; therefore, this category is called mixed. Together, these fatty acid categories represent the total fatty acid content of milk resulting in the following relationship:

Milkfat = glycerol + fatty acids (de novo + mixed + preformed)

Under normal circumstances, 25% of the fatty acids are only synthesized de novo, 37.5% are in the mixed category, and 37.5% are in the preformed category (Woolpert, 2016). However, a 1 unit change in any fatty acid category results in about the same change in milk fat. For example, when milk with 3.8% fat has a decline in de novo content from 0.9 to 0.7, the fat content will decrease to 3.6%. As diet changes, these proportions of each category also change suggesting that they can provide insight in cow performance.

The balance of de novo fatty acids and preformed fatty acids in milk changes dramatically during the post calving transition period. After calving the preformed fatty acids are a high proportion of the total fatty acids in the milk fat (e.g., 50% or higher) and the de novo fatty acids are about 20% of the total fatty acids. When cows come into positive energy balance, the portion of de novo and preformed fatty acids in the milk should stabilize. The point at which they stabilize and move up and down during the remainder of lactation will be function of management practices (e.g., stocking density) and feed nutritional characteristics and quality.

Another feature of milk fat from ruminants is that it is highly saturated. Double bonds in milk fat come from two sources. First, there can be a high percentage of unsaturated fat reaching the mammary cell. This is usually detrimental as native unsaturated fat that escapes the rumen suggests incomplete biohydrogenation of dietary fat. Incomplete biohydrogenation can result from a heavy load of unsaturated fat, extremely high passage rates, or from impaired rumen function. Another scenario is that unsaturated fatty acids (particularly C18:1) may be provided

in the diet as rumen protected fatty acids. Being rumen protected, these unsaturated fatty acids have minimal effects on the rumen environment but can still affect the mammary cell when high levels are incorporated into milk fat.

Unsaturated fatty acid can also be produce in the mammary cells by the enzyme stearyl CoA desaturase. This enzyme converts C18:0 fatty acid (stearic acid) to C18:1 cis-9 fatty acid (oleic acid). This enzyme may play a role in maintaining the fluidity of milk fat. Milk fat needs to have a melting point lower than the body temperature of the cow for secretion.

The issue of chain length (carbons/fatty acids) and degree of unsaturation (double bonds per fatty acid) appears to be a fluidity issue. If chain length increases without a corresponding increase in double bonds, the fluidity of the milk fat would decrease. Likewise, an increase in double bonds decreases fluidity. For example, if there are a lot of unsaturated preformed fatty acids, de novo synthesis will be reduced (Barbano and Sherbon,1980). Remember that melting point goes down with the addition of double bonds and with shorter chain lengths.

Recently, 68 Holstein herds were analyzed for milk fatty acid composition by Cornell University. Some of these herds also conducted a TMR fatty acid analysis through Cumberland Valley Analytical Services. These are not randomly selected herds as they were submitted by herd consultants and nutritionists who were interested in learning more about the milk fatty acid profile of their herds. The mean, min and max values for 68 bulk tank samples are in Table 1.

References

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Table 1. Mean, min and max milk composition values for 68 Holstein herds.

	Protein	Fat	FA	de novo	Mixed	Preformed	carbon #	DB/FA	Fluidity
	g/100 g milk						FA CL	FA Unsat	C/DBL
Mean	3.06	3.64	3.43	0.81	1.30	1.31	14.65	0.32	46.07
Min	2.80	3.09	2.89	0.65	1.08	1.13	14.42	0.28	42.58
Max	3.34	4.16	3.94	1.00	1.52	1.57	14.90	0.35	52.08

Table 1 demonstrates that the large range in milk fat (1.07 units) is accompanied with large ranges in the individual fatty acid composition (*de novo*, 0.35; mixed, 0.44; preformed, 0.44 units). Milk protein content had a much narrower range (0.54 units) compared to total milk fat. This suggests that there are effects on milk fat that are much larger than the effects on milk protein. The column labeled Carbon # (FA CL; fatty acid chain length) refers to the average number of carbons per fatty acid. Fatty acid chain length is very much dependent on proportions of fatty acids in each category. As the proportion of preformed fatty acids increases, the chain length will increase. Since longer fatty acids have higher melting points, a longer chain length will decrease fluidity. The range in Table 1 for Carbon # was 14.42 to 14.90. DB/FA (FA Unsat) refers to the number of double bonds per fatty acid. As double bonds are added to fatty acids, melting point decreases which increases fluidity. The range in Table 1 for DB/FA was 0.28 to 0.35. Because there is an inverse relationship between carbon # and DB/FA relative to fluidity, a fluidity index of (carbon #)/(DB/FA) has been developed. Across the dataset, fat percent increases as the fluidity index increases (fat percent = 0.84 + .06*fluidity; R^2 = 0.30).

As a point of reference, Table 2 contains the milk component values for an example herd with very good milk composition based on the high level of *de novo* milk fatty acids and typical ratios of mixed and preformed fatty acids.

Table 2. An example herd with excellent milk components.

Protein	Fat	FA	de novo	Mixed	Preformed	carbon #	DB/FA	Fluidity
g/100 g milk						FA CL	FA Unsat	C/DBL
3.13	3.89	3.75	0.87	1.41	1.46	14.50	0.28	52

The 68 herds fall into several categories:

High levels of dietary unsaturated fat affecting the mammary gland. CLA (C18:2 *trans*-10:*cis*-12) has been shown to be a potent inhibitor of *de novo* milk fat synthesis in the mammary cell. These herds will have normal rumen function, but *de novo* synthesis will be down regulated. In this example herd (Table 3), fat is depressed to 3.46% but milk protein is near normal (3.1%). This suggests that the rumen is producing sufficient metabolizable protein to support high levels of milk protein production. However, *de novo* fatty acid synthesis is impaired. In this case, *de novo* milk fat synthesis is low (0.72 vs 0.78). The preformed fatty acids were high, as there was added fat in the diet. The increased percentage of preformed fatty acids led to increased average chain length; however, the number of double bonds is exceedingly high resulting in a more fluid fat. The fluidity index is low which is suggesting an imbalance in chain length and unsaturated fat.

Table 3. Example herd exhibiting normal milk protein, low milk fat, low de novo fatty acids, and a high level of unsaturated fat.

Protein	Fat	FA	de novo	Mixed	Preformed	carbon #	DB/FA	Fluidity
g/100 g milk						FA CL	FA Unsat	C/DBL
3.10	3.46	3.23	0.72	1.20	1.31	14.90	0.34	43
Expected			0.78	1.23	1.23			

1) **High levels of dietary unsaturated fat affecting both the rumen and the mammary gland**. Unsaturated fat can impair ruminal fiber digestion which will reduce ruminal protein production in addition to providing a high level of unsaturated fat directly to the mammary gland. In the example herd (Table 4), roasted soybeans were included in the diet resulting in an abnormally high level of dietary unsaturated fat. Both protein and fat content of the milk are depressed with a lowered *de novo* milk fatty acid synthesis (0.70 versus 0.80 g/100 g milk). Added fat in the diet is raising both the mixed and preformed categories. With the lowered *de novo* synthesis, fatty acid chain length is longer but again the amount of double bonds is higher than expected given this increase in chain length with a low fluidity index.

Table 4. Example herd with low milk protein, low milk fat, low *de novo* fatty acids and high level of unsaturated fatty acids.

Protein	Fat	FA	de novo	Mixed	Preformed	carbon #	DB/FA	Fluidity
g/100 g milk						FA CL	FA Unsat	C/DBL
2.94	3.57	3.34	0.70	1.27	1.37	14.82	0.34	44
Expected			0.80	1.27	1.27			

1) **A shortage of de novo milk fatty acids without a high degree of unsaturated fat**. These herds appear normal except that the milk fat is depressed. In the example herd (Table 5), fat is slightly depressed while protein and amount of unsaturated fatty acids are near normal. Herds such as this appear to have a shortage of substrate for *de novo* synthesis rather than an inhibition of *de novo* synthesis. It is widely recognized that acetate and butyrate are the building blocks of *de novo* fat synthesis in the mammary gland with much of the focus on acetate. However, butyrate may play a more important role than previously recognized. For example, 36 mole% of triglycerides contained C4 (butyrate) or C6 (butyrate + acetate) (Jensen, 2002). All the C4 and 90% of the C6 fatty acids were on the *sn*-3 position (the third leg of the triglyceride). Numerous rumen microflora produce butyrate, however, a primary substrate used in producing butyrate may be sugar (glucose and sucrose).

Table 5. An example herd with low fat with near normal protein and low amount of unsaturated fatty acids.

Protein	Fat	FA	de novo	Mixed	Preformed	carbon #	DB/FA	Fluidity
g/100 g milk						FA CL	FA Unsat	C/DBL
3.06	3.50	3.31	0.78	1.27	1.25	14.71	0.31	48
Expected			0.80	1.27	1.27			

1) **Excessive levels of palm fat in the diet**. High levels of palm fat (C16:0) in the diet can mask other fat production issues. In the herd shown in Table 6, protein and total fat are slightly

reduced. In this example, the level of mixed fatty acids is high (1.46 vs 1.30 g/100g milk). If the mixed fatty acids were not elevated, the actual fat content would be closer to 3.45% as opposed to the observed 3.62%. For most corn based diets, C16:0 represents about 20% of the total fatty acids. In this herd, the TMR fatty acid report (Figure 1) showed 35% of the total fatty acids were C16:0. Clearly, a C16:0 supplemental product is being added to the diet. Milk fatty acid composition for this herd suggests that more *de novo* synthesis is needed, probably dependent on sugar availability for ruminal butyrate synthesis, along with more total energy to spare the preformed fatty acids. Since the degree of unsaturation is low, adding more corn would be appropriate.

Table 6. Example herd with high levels of C16 fatty acids due to supplemental palm fat.

Protein	Fat	FA	de novo	Mixed	Preformed	carbon #	DB/FA	Fluidity
g/100 g						FA CL	FA Unsat	C/DBL
2.96	3.62	3.42	0.80	1.46	1.16	14.64	0.29	51
Expected			0.82	1.30	1.30			

Figure 1. TMR fatty acid levels for an example herd with high levels of palmitic acid. Typical corn based diets usually contain 20% of the fatty acids as C16:0 with no supplemental fat.

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