ROLE OF 16- AND 18-CARBON FATTY ACIDS IN DAIRY RATIONS

A. L. Lock and J. de Souza
Department of Animal Science
Michigan State University

INTRODUCTION

In most Federal Milk Market Orders milk fat and protein yield are the major contributors to the price that producers receive for milk. The addition of supplemental fatty acid (FA) sources to diets is a common practice in dairy nutrition to increase dietary energy density and to support milk production and solids yield. The ability to understand and model FA, the effects of individual FA, and different FA supplements on production parameters has direct impact on dairy industry recommendations and the usefulness of FA supplementation strategies. The emphasis of the current paper is on biological processes and quantitative changes during the metabolism of FA in the rumen and the effect this has on FA availability to the dairy cow, the digestibility of these FA, and their overall impact on performance and energy partitioning. We will focus on recent research supplementing palmitic acid (C16:0) and stearic acid (C18:0)-enriched supplements, on feed intake, milk production, milk composition, and energy partitioning.

Lipid Metabolism in The Rumen and Mammary Gland

As well as being derived from specific supplements, FA in the dairy cow's diet are also present in forages and concentrates. Each feed/fat source is composed of a different mix of individual FA. The majority of FA in dairy cow diets contain 16 and 18-carbons. Generally, most cereal grains and seeds contain a high concentration of linoleic acid (C18:2 n-6), whereas linolenic acid (C18:3 n-3) is typically the predominant FA in forage sources. For example, corn, cottonseed, safflower, sunflower, and soybean oils are high in C18:2 n-6, whereas linseed is high in C18:3 n-3. Unsaturated FA are toxic to many rumen bacteria, thus an extensive metabolism of dietary lipids occurs in the rumen that has a major impact on the profile of FA available for absorption and tissue utilization (Palmquist et al., 2005). The two major processes that occur are hydrolysis of ester linkages in lipids found in feedstuffs and the biohydrogenation of unsaturated FA. Biohydrogenation of unsaturated FA results in the conversion of unsaturated FA to saturated FA, mainly C18:0, through a series of biohydrogenation intermediates (conjugated C18:2 and *trans* C18:1 FA). The major substrates are 18:2 n-6 and 18:3 n-3 and the rate of rumen biohydrogenation is in the range of 70-95% and 85-100%, respectively (Jenkins et al., 2008); thus C18:0 is the predominant FA available for absorption by the dairy cow under typical feeding situations (Bauman and Lock, 2006). A series of recent in vitro studies concluded that biohydrogenation occurs to enable rumen bacteria to survive the bacteriostatic effects of unsaturated FA, and that the toxicity of unsaturated FA is probably mediated via metabolic effects rather than disruption of membrane integrity. Furthermore, it appears that the degree of toxicity of different unsaturated FA varies for individual ruminal bacteria species; all the main species that

comprise the ruminal cellulolytic bacteria appear vulnerable to inhibition by unsaturated FA (Maia et al., 2007, 2010).

FA supplements are often used as a means to increase the energy density of the diet and many of these are referred to as inert. In this case inertness simply means that the FA supplement has minimal affects on rumen fermentation. Although deemed inert at the level used, they can still be hydrolyzed, if a triglyceride, or biohydrogenated, if unsaturated. Often, Calcium-salts of palm FA or canola are referred to as 'protected'. However, these are not protected from rumen biohydrogenation, but rather are considered to be ruminally inert with regard to their effects on the microbial population (Palmquist, 2006).

Lipids in milk are primarily in the form of triglycerides (98%) with phospholipids and sterols accounting for 1.0 and 0.5 % of total lipids, respectively. Bovine milk is extremely complex and contains about 400 FA, a large proportion of which are derived from lipid metabolism in the rumen (Jensen, 2002). Milk FA are derived from 2 sources; <16 carbon FA from de novo synthesis in the mammary gland and >16 carbon FA originating from extraction from plasma. 16-carbon FA originate from either de novo or preformed sources. Substrates for de novo synthesis are derived from ruminal fiber digestion and dietary FA supply preformed FA for direct incorporation into milk fat (Palmquist, 2006). Microbial synthesis of branched and odd-chained number FA in the rumen and absorption of biohydrogenation intermediates also contribute to the diversity of FA secreted in milk fat. Under typical conditions, about half of the FA in milk are synthesized de novo, 40 to 45 % originate from FA in the diet, and less than 10% are derived from mobilization of adipose tissue (Palmquist and Jenkins, 1980). However, nutrition can substantially alter the balance between mammary de novo FA synthesis and uptake of preformed FA. C16:0, C18:0 and *cis*-9 C18:1 are the major FA in milk fat. The relatively high melting point of C16:0 and C18:0 requires the production of de novo synthesized FA or the conversion of C16:0 and C18:0 to *cis*-9 C16:1 and *cis*-9 C18:1, respectively, in the mammary gland in order to maintain fluidity.

Overall Impact of Fa Supplements

There is a wide range of FA supplements available for lactating dairy cattle. For example, Calcium-salts of free FA and prilled saturated free FA are two common types of supplements used in the dairy industry and they differ in FA content and FA profile. Calcium-salt supplements typically contain 80-85% FA and these typically provide approximately 50% saturated and 50% unsaturated FA. By comparison prilled saturated free FA contain approximately 99% FA which are approximately 90% saturated, 10% unsaturated. A summary of the FA profile of some commonly used supplements is provided in Table 1. Although in general FA supplementation has been shown to increase milk yield, milk fat yield, and the efficiency of milk production, great variation has been reported in production performance for different FA types, and indeed the same supplement across different diets and studies. This is evident in a meta-analysis examining the effect of FA supplementation to diets of dairy cows (Rabiee et al., 2012). In general milk production and milk fat % and yield increased, DMI and milk protein %

decreased, and milk protein yield was not affected by FA supplementation. There was a wide range of responses (~5 standard deviations) for all variables, indicating varied and marked biological effect of the different FA supplements (Rabiee et al., 2012).

Table 1. Fatty acid composition of common fat supplements (Data from our laboratory).

Fatty Acid, g/100 g	Tallow	Ca-salt PFAD	Saturated free FA	C16:0-enriched
C14:0	3.0	2.0	2.7	1.6
C16:0	24.4	51.0	36.9	89.7
C18:0	17.9	4.0	45.8	1.0
C18:1	41.6	36.0	4.2	5.9
C18:2	1.1	7.0	0.4	1.3

Utilizing a larger data set than Rabiee et al. (2012), we recently performed a meta-analysis of production responses to commercially available FA supplements (Boerman and Lock, 2014a). Available data were collected from 133 peer-reviewed publications of which 88 met our selection criteria, comprising 159 treatment comparisons. Calcium-salts of palm FA distillate (PFAD; n=73), saturated prilled FA (PRILLS; n=37), and tallow (n=49) supplemented at ≤ 3% diet DM were compared to non FA supplemented diets used as controls. Treatment comparisons were obtained from either randomized design (n=99) or crossover/Latin square design experiments (n=60). Preliminary results from the meta-analysis are shown in Figure 1.

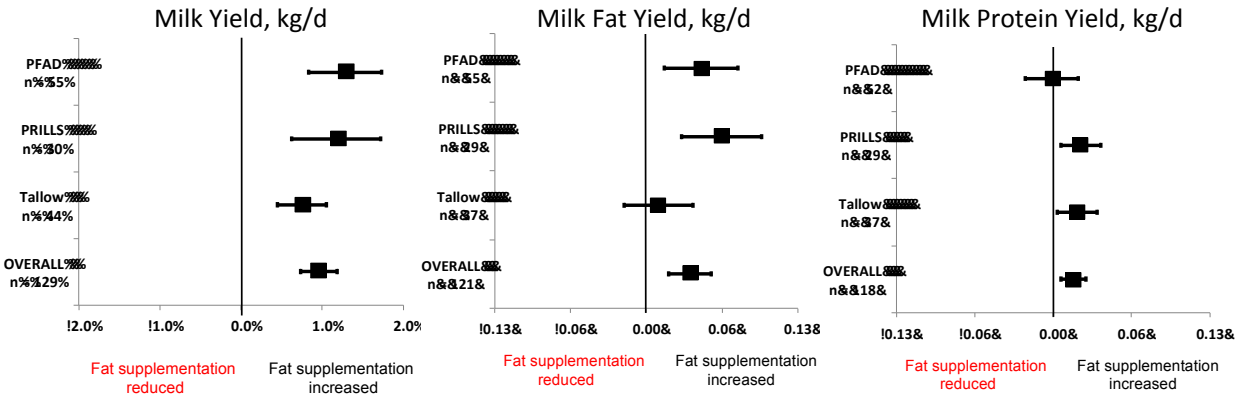

Figure 1.Effect of commercially available FA supplements on yield of milk, milk fat, and milk protein (Boerman and Lock, 2014a). All data reported in peer-reviewed journals in which FA supplements were included at ≤ 3% diet DM compared to control with no added FA supplement. All studies had to have measurements of variance reported. PFAD – calcium salts of palm FA distillate (~ 50% 16:0, ~ 50% unsaturated 18-carbon FA); PRILLS – saturated FA prills (> 80% saturated FA [16:0 and/or 18:0]); Tallow – animal fat labeled as tallow (~ 50% 16:0 and 18:0, ~ 45% 18:1). Data analyzed using *Comprehensive Meta-Analysis (CMA) version 2.0 (Biostat, Englewood, NJ), calculating difference between FA supplemented and control diets using a random effects model.*

Overall, FA supplementation increased yield of milk and milk components and reduced DMI. However type of supplement influenced response with PRILLS not reducing DMI, tallow having no effect on milk fat yield, and PFAD having no effect on milk protein yield. It is important to note that the majority of the studies reported in Figure 1 simply compared a single commercial FA supplement with a non FA supplemented control diet. This makes direct comparisons between different FA supplements difficult to interpret and importantly provide accurate answers to commonly asked questions (by farmers and nutritionists) as to which are the best FA supplements to use. There are limited reports in the published literature that have undertaken direct comparisons between different commercially available FA supplements. Results from the meta-analysis also suggest that responses to FA supplements interact with other dietary components, and this should be examined further.

Impact of Supplemental 16- And 18-Carbon Fa on Fa Digestibility

Under typical feeding situations, C18:0 is the predominant FA available for absorption by the dairy cow, regardless of the diet fed. As result, this FA has an important impact on total FA digestibility as recently observed in a recent meta-analysis and meta-regression examining the intestinal digestibility of long-chain fatty acids in lactating dairy cows (Boerman et al., 2015a). We observed a negative relationship between the total flow and digestibility of FA (Figure 2A). Furthermore, the decrease in total FA digestibility appears to be driven by the digestibility of C18:0 because a negative relationship between the duodenal flow and digestibility of C18:0 was also was detected (Figure 2B).

The exact mechanisms for the reduction in digestibility are not understood; however, potential causes include limits in lysolecithin or competition for absorption sites (Drackey, 2000). Lysolecithin also acts as an amphiphile (substance with both water and lipid-loving capacity) and further increases the solubility of saturated FA (Freeman, 1969). During FA digestion in the small intestine, bile secretions supply bile salts and lecithin, and pancreatic secretions provide enzymes to convert lecithin to lysolecithin and bicarbonate to raise the pH. Lysolecithin is an emulsifier compound and together with bile salts desorb FA from feed particles and bacteria, allowing the formation of micelles, which is critical for absorption (Lock et al., 2005). Once micelles are formed they facilitate transfer of water-insoluble FA across the unstirred water layer of intestinal epithelial cells, where the FA and lysolecithin are absorbed. Additional research to understand the observed reduction in C18:0 digestibility and how this may be overcome or improved is required.

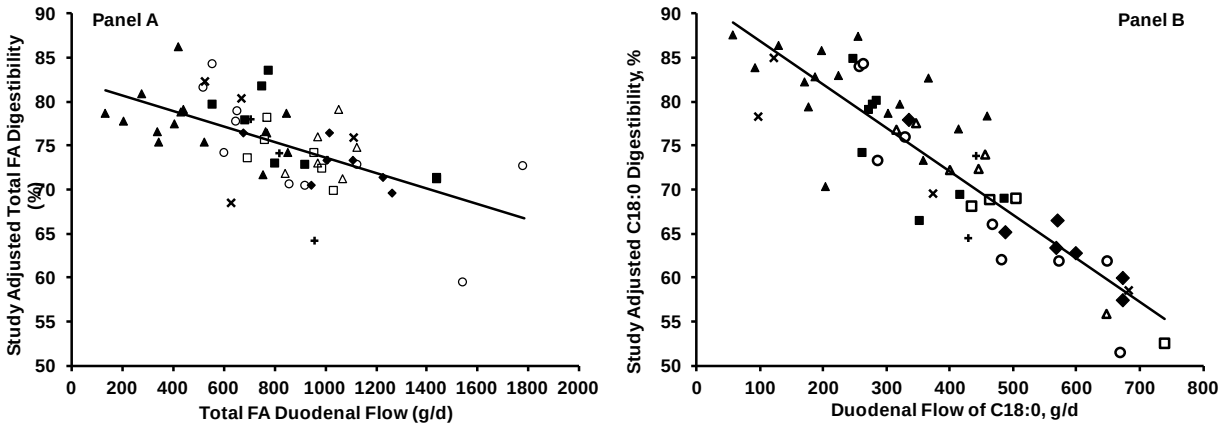

Figure 2. Relationship between study adjusted total FA intestinal digestibility and total FA duodenal flow (Panel A) and study adjusted C18:0 intestinal digestibility and duodenal flow of C18:0 (Panel B). Results from a meta-analysis using 15 published studies that measured duodenal flow and intestinal digestibility of fatty acids in dairy cows (Boerman et al., 2015a). Control treatments represented by black triangles; animal-vegetable fat treatments represented by black diamonds; calcium salt treatments represented by black squares; tallow treatments represented by open circles; vegetable oil treatments represented by open triangles; seed meal treatments represented by open squares; whole seed treatments represented by black addition sign; and other treatments represented by black multiplication sign.

Our recent FA digestibility research has utilized and focused on C16:0 and C18:0-enriched supplements. Of particular importance, Boerman et al. (2014b) fed increasing levels of a C18:0-enriched supplement (85% C18:0) to dairy cows and observed no positive effect on production responses, which was likely associated with the pronounced decrease in total FA digestibility as FA intake increased (Figure 3A). Similarly, de Souza et al. (2015) fed increasing levels of a C16:0-enriched supplement (87% C16:0) to dairy cows and even though a positive effect was observed on production response up to 1.5% diet dry matter, we observed a decrease in total FA digestibility as FA intake increased (Figure 3B). Considering the results presented in Figure 3, given that the range on FA intake is similar across both studies, the decrease in total FA digestibility is more pronounced when there is increased intake/rumen outflow of C18:0 rather than C16:0, similar to our observations in Figure 2.

To further understand what factors influence FA digestibility, we recently utilized a random regression model to analyze available individual cow data from 5 studies that fed a C16:0-enriched supplement to dairy cows (unpublished results). We observed that total FA digestibility was negatively impacted by total FA intake, but positively influenced by the intake of *cis*-9 C18:1. This suggests that a combination between 16-carbon and unsaturated 18-carbon FA may improve FA digestibility, but reason for this effect needs to be further determined.

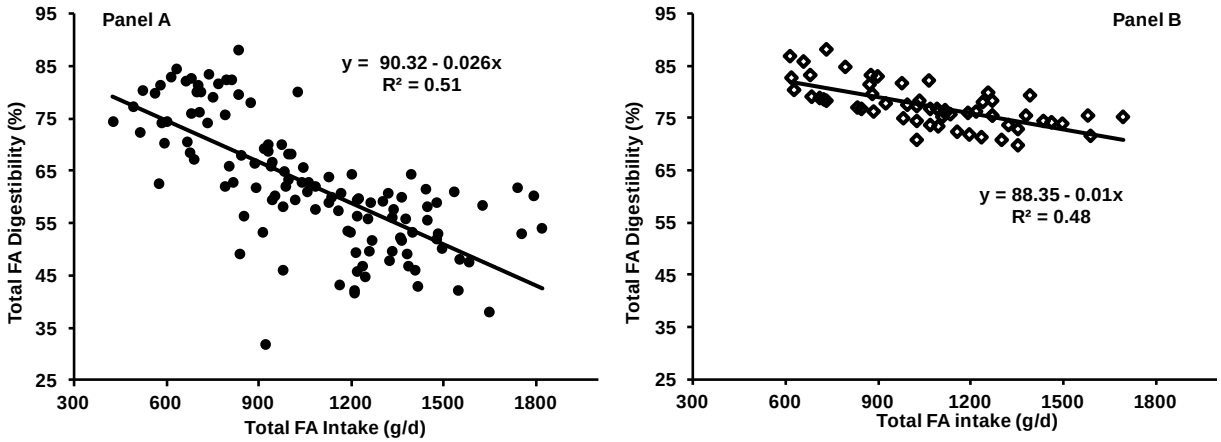

Figure 3. Relationship between total FA intake and total FA digestibility of dairy cows supplemented with either a C18:0-enriched supplement (Panel A) or a C16:0-enriched supplement (Panel B). Results in Panel A utilized 32 mid-lactation cows receiving diets with increasing levels (0 to 2.3% dry matter) of a C18:0-enriched supplement (85% C18:0) in a 4 X 4 Latin square design with 21-d periods (Boerman and Lock, 2014b). Results in Panel B utilized 16 mid-lactation cows receiving diets with increasing levels (0 to 2.25% dry matter) of a C16:0-enriched supplement (87% C16:0) in a 4 X 4 Latin square design with 14-d periods (De Souza et al., 2015).

Impact of Supplemental 16- And 18-Carbon Fa on Production Responses

In the 1960's Steele and co-workers performed a series of studies using relatively pure sources of C16:0 and C18:0 and and their findings suggested that C16:0 supplementation induces a higher milk fat response (concentration and yield) as compared to C18:0 supplementation. More recent work from Enjalbert et al (1998) suggests that the uptake efficiency of the mammary gland is higher for C16:0 than for C18:0 and *cis*-9 C18:1. We recently carried out a series of studies examining the effect of individual saturated FA on production and metabolic responses of lactating cows (Lock et al., 2013, Piantoni et al., 2013, Rico et al., 2014, Piantoni et al., 2015). These results indicate that C16:0 supplementation has the potential to increase yields of milk and milk fat as well as the conversion of feed to milk, independent of production level when it was included in the diet for soyhulls or C18:0 (Table 2).

Rico et al. (2013) fed increasing levels of a C16:0-enriched supplement (87% C16:0) to dairy cows and observed a quadratic response with a positive effect on milk fat yield, 3.5% fat-corrected milk and feed efficiency up to 1.5% diet DM (Table 3). Furthermore, we recently utilized a random regression model to analyze available individual cow data from 10 studies that fed a C16:0-enriched supplement to dairy cows (unpublished results). We observed that energy partitioning toward milk was increased linearly with C16:0 intake, as a result of a linear increase in milk fat yield and 3.5% fat-corrected milk with increasing intake of C16:0.

Table 2. Summary of DMI, milk production and composition, body weight, and BCS for cows supplemented with C16:0 and C18:0 supplements. The C16:0 supplement contained ~ 99% C16:0 and the C18:0 supplement contained ~ 98% C18:0.

	Piantoni et al. (2013)[1]			Piantoni et al. (2015)[2]			Rico et al. (2014)[3]		
Variable	**Control**	**C16:0**	**SEM**	**Control**	**C18:0**	**SEM**	**C16:0**	**C18:0**	**SEM**
DMI, kg/d	27.8	27.8	0.54	25.2^{n}	26.1^{m}	0.42	32.1	32.3	0.44
Milk yield, kg/d	44.9^{b}	46.0^{a}	1.7	38.5^{n}	40.2^{m}	0.71	46.6	45.8	2.02
Fat yield, kg/d	1.45^{b}	1.53^{a}	0.05	1.35^{n}	1.42^{m}	0.03	1.68^{y}	1.59^{z}	0.05
Milk fat, %	3.29^{b}	3.40^{a}	0.11	3.60	3.59	0.12	3.66^{y}	3.55^{z}	0.09
Protein yield, kg/d	1.38	1.41	0.04	1.14^{n}	1.19^{m}	0.02	1.50	1.49	0.05
Milk Protein %	3.11	3.09	0.05	3.00	2.99	0.05	3.24	3.29	0.05
3.5% FCM	42.9^{b}	44.6^{a}	1.35	38.6^{n}	40.5^{m}	0.76	47.5^{y}	45.6^{z}	1.64
3.5% FCM/DMI	1.54^{b}	1.60^{a}	0.03	1.53	1.55	0.04	1.48^{y}	1.40^{z}	0.05
Body weight, kg	722	723	14.7	727	730	12.8	720	723	13.6
BCS	2.99	2.93	0.15	2.67	2.67	0.11	2.93^{z}	2.99^{y}	0.11

[1]Treatments were either a control diet (with 2% of diet DM as added soyhulls) or a C16:0-supplemented diet (with 2% of diet DM as C16:0). Means within a row with different superscripts ($^{a, b}$) differ ($P < 0.05$).

[2]Treatments were either a control diet (with 2% of diet DM as added soyhulls) or a C18:0-supplemented diet (with 2% of diet DM as C18:0). Means within a row with different superscripts ($^{m, n}$) differ ($P < 0.05$).

[3]Treatments were either a C16:0-supplemented diet (with 2% of diet DM as C16:0) or a C18:0-supplemented diet (with 2% of diet DM as C18:0). Means within a row with different superscripts ($^{y, z}$) differ ($P < 0.05$).

Piantoni et al. (2015) reported that C18:0 increased DMI and yields of milk and milk components, with increases more evident in cows with higher milk yields, indicating that there was significant variation in response. Reasons why only higher yielding cows responded more positively to C18:0 supplementation than lower yielding cows remains to be determined. However, when we directly compared C16:0 and C18:0 supplementation the yield of milk fat and 3.5% FCM increased with C16:0 regardless of level of milk production (Table 2, Rico et al., 2014). In a recent dose response study with mid lactation cows feeding a C18:0-enriched supplement (85% C18:0) increased DMI but had no effect on the yields of milk or milk components when compared to non-FA supplemented control diet (Table 4), which is probably associated with the decrease in FA digestibility (Figure 3A, Boerman and Lock, 2014b).

There is mechanistic data to support the concept that individual FA can impact milk fat synthesis differently. Hansen and Knudsen (1987) utilized an in vitro system and reported that C16:0 stimulated de novo FA synthesis and incorporation into triglycerides whereas other FA were either neutral or inhibitory. In addition, there were only minor differences in the esterification efficiency into triglycerides of various FA, except for C16:0, which was a better substrate than the other FA tested. These results in association with the digestibility results suggest that C16:0-enriched supplement improve performance of dairy cows, while understanding factors that affect the digestibility of C18:0 with increasing intake/duodenal flow may allow the development of strategies to overcome this possible limitation.

Table 3. DMI, milk production and composition, body weight, and BCS for cows supplemented with increasing levels of a C16:0-enriched supplement (Rico et al., 2013). The C16:0 supplement contained 87% C16:0.

	C16:0 supplementation, % diet DM					
Variable	**0%**	**0.75%**	**1.50%**	**2.25%**	**SEM**	***P*-value**
DMI, kg/d	28.8	28.8	28.6	27.4	0.83	0.05
Milk yield, kg/d	43.7	43.5	44.5	42.5	1.73	0.06
Fat yield, kg/d	1.63	1.69	1.78	1.70	0.09	0.01
Milk Fat, %	3.78	3.88	4.01	4.03	0.17	0.01
Protein yield, kg/d	1.36	1.36	1.40	1.32	0.06	0.08
Milk Protein, %	3.17	3.15	3.18	3.16	0.07	0.32
3.5% FCM, kg/d	45.3	46.1	48.0	45.9	1.91	0.02
3.5% FCM/DMI	1.57	1.60	1.68	1.68	0.07	0.21
Body weight, kg	703	705	701	701	25.7	0.76
BCS	2.66	2.48	2.71	2.84	0.05	0.94

Table 4. DMI, milk production and composition, body weight, and BCS for cows supplemented with increasing levels of a C18:0-enriched supplement (Boerman and Lock, 2014b). The C18:0 supplement contained 85% C18:0.

	C18:0 supplementation, % diet DM				**SEM**	***P*-value**
Variable	**0%**	**0.80%**	**1.50%**	**2.30%**		
DMI, kg/d	28.5	29.1	29.6	30.0	0.61	0.13
Milk Yield, kg/d	38.3	38.6	38.2	37.8	1.65	0.51
Fat Yield, kg/d	1.43	1.40	1.40	1.42	0.04	0.61
Fat, %	3.79	3.72	3.74	3.82	0.08	0.29
Protein Yield, kg/d	1.33	1.33	1.32	1.30	0.05	0.49
Protein, %	3.49	3.50	3.48	3.49	0.05	0.91
3.5% FCM/DMI	39.8	39.4	39.3	39.3	1.40	0.77
FCM/DMI	1.43	1.39	1.35	1.33	0.04	0.03
Body weight, kg	738	739	735	737	12.0	0.58
BCS	3.44	3.40	3.39	3.42	0.08	0.37

Supplemental Fat Interactions with Other Dietary Components

The composition of the basal diet can also be an important element of production responses to FA supplementation. In high producing dairy cows an interaction was observed between forgage:concentrate ratio and response to supplemental FA (Weiss and Pinos-Rodriguez, 2009). In high-forage diets increased energy intake from supplemental saturated FA (mixture of C16:0 and C18:0) was directed mostly to body reserves, whereas in low-forage diets the increased energy intake from the saturated FA supplement was directed mostly to milk production. Using lower producing cows Grum et al. (1996) compared diets at 2 different forage:concentrate ratios either without or with added saturated FA (mixture of C16:0 and C18:0). At both forage:concentrate levels supplemental saturated FA increased milk fat concentration and yield, whereas saturated FA supplementation had opposing effects on DMI when supplemented in the low and high forage:concentrate diets. In early lactation cows, van Knegsel et al. (2007) fed either high

FA or high starch diets with the same concentrate to forage ratio (40:60). Additional FA in the high FA diet were provided by Ca-salts of palm FA and palm oil. Cows fed the high FA diet partitioned more energy to milk than cows fed the high starch diet and had a higher milk fat yield. No differences were found for energy retained as body protein, but energy mobilized from body fat tended to be higher in cows fed the lipogenic diet (van Knegsel et al., 2007).

In a recent study using high producing post-peak dairy cows we fed either a high fiber and FA diet (HFF) containing a 50:50 ratio of forage to concentrate containing a C16:0-enriched supplement at 2.5% of diet DM or a high starch diet (HS) containing a 40:60 ratio of forage to concentrate (Boerman et al., 2015b). The two treatments resulted in similar apparent energy densities and intakes but the HS treatment partitioned more energy toward body gain whereas the HFF treatment partitioned more energy toward milk (Table 5). In established lactation, cows are usually in positive energy balance and the goals are to maximize milk and component yields and reduce excessive conditioning. We recently observed that reducing starch concentration (32 to 16% diet DM) reduced BW gain in late lactation cows and diminished the incidence of over conditioning, while supplementation with a C16:0-enriched supplement increased milk fat yield and fat-corrected milk (Garver et al., 2015). Further work is necessary, but higher fiber and FA diets (particularly diets supplemented with palmitic acid) may diminish the incidence of over conditioning in mid and late lactation cows.

CONCLUSION

The addition of supplemental FA to diets is a common practice in dairy nutrition to increase dietary energy density and to support milk production. Although in general FA supplementation has been shown to increase milk yield, milk fat yield, and the efficiency of milk production, great variation has been reported in production performance for different FA supplements, and indeed the same supplement across different diets and studies. Further work is required to characterize the sources of variation in response to FA supplementation. Just as we recognize that not all protein sources are the same it is important to remember that not all FA supplements are the same. The key is to know what FA are present in the supplement, particularly FA chain length and their degree of unsaturation. Once this information is known it is important to consider the possible effects of these FA on DMI, rumen metabolism, small intestine digestibility, milk component synthesis in the mammary gland, energy partitioning between the mammary gland and other tissues, and body condition. Interactions with other dietary components and the level of milk production are also important in determining the response to various FA supplements. The extent of these simultaneous changes along with the goal of the nutritional strategy employed will ultimately determine the overall effect of the supplemental FA, and the associated decision regarding their inclusion in diets for lactating dairy cows.

Table 5. Body weight, body condition score, and calculated energy values for cows fed a high fiber diet containing a palmitic acid-enriched supplement or a high starch diet containing a mixture of dry ground and high moisture corn (Boerman et al., 2015b).

	Treatments[1]			
Variable	**HFF**	**HS**	**SEM**	***P*-value[2]**
DMI, kg/d	26.9	27.4	0.38	0.02
3.5% FCM, kg/d	49.1	47.6	1.59	0.03
Change in BW, kg/d	0.33	0.78	0.10	0.003
Change in BCS, pt/28 d	- 0.01	0.24	0.03	0.001
Calculated energy values[3]				
Apparent NE_L of diet Mcal/kg	1.78	1.79	0.02	0.64
Milk, Mcal/d	32.8	32.6	1.05	0.05
Body Tissue Gain, Mcal/d	1.95	4.90	0.58	0.001
Maintenance, Mcal/d	10.6	10.7	0.17	0.02
Partitioning				
Milk, %	72.8	67.9	1.11	< 0.001
Body Tissue Gain, %	4.03	10.1	1.16	0.001
Maintenance, %	23.2	22.0	0.43	0.01

[1] Treatments were either a high fiber and FA diet (HFF) containing a 50:50 ratio of forage to concentrate containing a palmitic acid-enriched supplement at 2.5% of diet DM or a high starch diet (HS) containing a 40:60 ratio of forage to concentrate containing a mixture of dry ground and high moisture corn.
[2] *P*-value associated with treatment differences (HFF vs. HS; Trt).
[3] From the sum of milk energy output, maintenance energy calculated from metabolic BW, and body energy gain divided by DMI for each cow on each diet throughout the 28-d period.

REFERENCES

Bauman, D.E., and A.L. Lock. 2006. Concepts in lipid digestion and metabolism in dairy cows. Pages 1-14. In: Proc. Tri-State Dairy Nutr. Conf. Available at: http://tristatedairy.osu.edu/

Boerman, J.P. and A.L. Lock. 2014a. Feed intake and production responses of lactating dairy cows when commercially available fat supplements are included in diets: a meta-analysis. J. Dairy Sci. 97 (E-Suppl. 1):319.

Boerman, J.P. and A.L. Lock. 2014b. Milk yield and milk fat responses to increasing levels of stearic acid supplementation of dairy cows. J. Dairy Sci. 97 (E-Suppl. 1):840.

Boerman, J.P., J.L. Firkins, N. St-Pierre, and A.L Lock. 2015a. Intestinal digestibility of long chain fatty acids in dairy cows: a meta-analysis and meta-regression. J. Dairy Sci. In Press: http://www.journalofdairyscience.org/inpress.

Boerman, J.P, S.B. Potts, M.J. VandeHaar, and A.L. Lock. 2015b. Effects of partly replacing dietary starch with fiber and fat on milk production and energy partitioning. J. Dairy Sci. 98:7264–7276

de Souza, J., J.E. Rico, C.L Preseault, M.S. Allen, and A.L. Lock. 2015. Total-tract fatty acid digestibility responses to increasing levels of palmitic acid supplementation of dairy cows receiving low- and high-fat diets. J. Dairy Sci. 98 (E-Suppl. 1):867.

Drackley, J.K. 2000. Lipid Metabolism. Pp. 97-119 in Farm Animal Metabolism and Nutrition. (ed. J. P. F. D'Mello). CABI Publishing, New York, NY.

Enjalbert, F., M.C. Nicot, C. Bayourthe, and R. Moncolon. 1998. Duodenal infusions of palmitic, stearic or oleic acids differently affect mammary gland metabolism of fatty acids in lactating dairy cows. J. Nutr. 128:1525–1532.

Freeman, C.P. 1969. Properties of fatty acids in dispersions of emulsified lipid and bile salt and the significance of these properties in fat absorption in the pig and the sheep. British J. Nutr. 23:249-263.

Garver, J.L., J. de Souza, M.J. VandeHaar, and A.L. Lock. 2015. Effects of including supplemental fat in low and high starch diets on milk production and energy partitioning. J. Dairy Sci. 98 (E-Suppl. 1):552.

Hansen, H., and J. Knudsen. 1987. Effect of exogenous long-chain fatty acids on lipid biosynthesis in dispersed ruminant mammary-gland epithelial cells - esterification of long-chain exogenous fatty acids. J. Dairy Sci. 70:1344–1349.

Jenkins, T.C., R.J. Wallace, P.J. Moate, and E.E. Mosley. 2008. Board-Invited Review: Recent advances in biohydrogenation of unsaturated fatty acids within the rumen microbial ecosystem. J. Anim. Sci. 86:397-412.

Jensen, R. 2002. The composition of bovine milk lipids: January 1995 to December 2000. J. Dairy Sci. 85:295–350.

Lock, A.L., C.L. Preseault, J.E. Rico, K.E. DeLand, and M.S. Allen. 2013. Feeding a C16:0-enriched fat supplement increased the yield of milk fat and improved conversion of feed to milk. J. Dairy Sci. 96:6650–6659.

Lock, A.L., K.J. Harvatine, I. Ipharraguerre, M. Van Amburgh, J.K. Drackley, and D.E. Bauman. 2005. The dynamics of fat digestion in lactating dairy cows: what does the literature tell us? Proc. of Cornell Nutrition Conference. P 83-94.

Maia, M.R.G., L.C. Chaudhary, C.S. Bestwick, A.J. Richardson, N. Mckain, T.R. Larson, I.A. Graham, and R.J. Wallace. 2010. Toxicity of unsaturated fatty acids to the biohydrogenating ruminal bacterium, Butyrivibrio fibrisolvens. BMC. Microbiol. 10:52.

Maia, M.R.G., L.C. Chaudhary, L. Figueres, and R.J. Wallace. 2007. Metabolism of polyunsaturated fatty acids and their toxicity to the microflora of the rumen. Antonie Van Leeuwenhoek. 91:303–314.

Palmquist, D.L. 2006. Milk fat: origin of fatty acids and influence of nutritional factors thereon. In: P. F. Fox and P. L. H. McSweeney (Eds.) Advanced Dairy Chemistry, Volume 2: Lipids, 3rd Edition. pp. 43-92. Kluwer Academic/Plenum Publishers, New York, USA.

Palmquist, D.L., A.L. Lock, K.J. Shingfield, and D.E. Bauman. 2005. Biosynthesis of conjugated linoleic acid in ruminants and humans. Pages 179-217. In: Advances in Food and Nutrition Research. Vol. 50. S.L. Taylor, (ed.). Elsevier Inc., San Diego, CA.

Palmquist, D.L., and T.C. Jenkins. 1980. Fat in lactation rations: Review. J. Dairy Sci. 63:1–14.

Piantoni, P., A.L. Lock, and M.S. Allen. 2013. Palmitic acid increased yields of milk and milk fat and nutrient digestibility across production level of lactating cows. J. Dairy Sci. 96:7143–7154.

Piantoni, P., A.L. Lock, and M.S. Allen. 2015. Milk production responses to dietary stearic acid vary by production level in dairy cattle. J Dairy Sci. 98:1938–1949.

Rabiee, A.R., K. Breinhild, W. Scott, H.M. Golder, E. Block, and I.J. Lean. 2012. Effect of fat additions to diets of dairy cattle on milk production and components: A meta-analysis and meta-regression. J. Dairy Sci. 95:3225–3247.

Rico, J.E., M.S. Allen, and A.L. Lock. 2014. Compared with stearic acid, palmitic acid increased the yield of milk fat and improved feed efficiency across production level of cows. J. Dairy Sci. 97:1057-1066.

Steele, W. 1969. The effects of dietary palmitic and stearic acids on milk yield and composition in the cow. J. Dairy Res. 369–373.

Steele, W., and J.H. Moore. 1968. The effects of a series of saturated fatty acids in the diet on milk-fat secretion in the cow. J. Dairy Res. 35:361–369.

van Knegsel, A.T.M., H. van den Brand, J. Dijkstra, W.M. van Straalen, M.J.W. Heetkamp, S. Tamminga, and B. Kemp. 2007. Dietary energy source in dairy cows in early lactation: energy partitioning and milk composition. J. Dairy Sci. 90:1467–1476.

Weiss, W.P., and J.M. Pinos-Rodríguez. 2009. Production responses of dairy cows when fed supplemental fat in low- and high-forage diets. J. Dairy Sci. 92:6144–6155.