### Feeding Blends of Fatty Acids for Transition Cows

Adam L. Lock<sup>1</sup>, Crystal M. Prom<sup>1</sup>, Arnulfo Pineda<sup>1</sup>, and Jonas de Souza<sup>2</sup>

<sup>1</sup>Department of Animal Science, Michigan State University

<sup>2</sup>Perdue Agribusiness

#### Introduction

Recently, the effects of individual fatty acids (FA) on digestibility, metabolism, and production responses of dairy cows has received attention. In fresh cows, the high metabolic demand of lactation and reduced DMI during the immediate postpartum period result in a state of negative energy balance. Approaches to increasing energy intake of postpartum cows include increasing starch content of the diet and supplementing FA to increase the energy density of the diet. However, feeding high starch diets that promote greater ruminal propionate production during early lactation could be hypophagic and therefore further reduce DMI and increase the risk of ruminal acidosis and displaced abomasum (Allen and Piantoni, 2013). Some authors suggest that caution should be exercised when using supplemental FA to increase the caloric density of diets in early lactation dairy cows, since a high lipid load may affect the endocrine system, feed intake, and increases the risk for metabolic disorders (Kuhla et al., 2016). However, just as we recognize that not all protein sources are the same, it is important to remember that not all FA or FA supplements are the same. We will briefly review the biological processes and quantitative changes during the metabolism of FA, the digestibility of these FA, and their overall impact on performance. Our emphasis in the current paper is on recent research supplementing palmitic (C16:0), stearic

(C18:0), and oleic (C18:1) acids on feed intake, nutrient digestibility, and milk production.

### Effect of Fatty Acids on NDF Digestibility

Changes in intake and digestibility of other nutrients, such as NDF, due to FA supplementation may affect positively or negatively the digestible energy value of any FA supplement. Weld and Armentano (2017) performed a meta-analysis to evaluate the effects of FA supplementation on DMI and NDF digestibilities of dairy cows. Addition of vegetable oil decreased NDF digestibility by 2.1 percentage units but did not affect DMI. Feeding saturated prilled supplements (combinations of C16:0 and C18:0) did not affect DMI, but increased NDF digestibility by 0.22 percentage units. Overall, the authors concluded that the addition of a fat supplement, in which the FA are 16-carbon or greater in length, has minimal effects on NDF digestibility, but the effect of C16:0-enriched supplements were not evaluated.

We recently utilized a random regression model to analyze available individual cow data from 6 studies whereby C16:0-enriched supplements were fed to dairy cows (de Souza et al., 2016). We observed that NDF digestibility was positively impacted by total C16:0 intake (Figure 1A) and DMI was not affected. This suggests that that the increase in NDF digestibility when C16:0-enriched supplements

<sup>&</sup>lt;sup>1</sup>Contact at: 474 S. Shaw Lane, 2265H Anthony Hall, East Lansing, MI 48824, (517) 802-8124, Email: allock@msu.edu.

are fed to dairy cows is not explained through a decrease in DMI. Additionally, when comparing combinations of C16:0, C18:0, and C18:1 in supplemental fat, we observed that feeding supplements containing C16:0 or C16:0 and C18:1 increased NDF digestibility compared with a supplement containing C16:0 and C18:0 (de Souza et al., 2018a).

With early lactation cows, Piantoni et al. (2015b) fed a saturated fat supplement (~ 40% C16:0 and 40% C18:0) and observed that fat supplementation increased NDF digestibility by 3.9% units in the low forage diet (20% fNDF) but had no effect in the high forage diet (26% fNDF). When evaluating the effects of timing of C16:0 supplementation (PA) on performance of early lactation dairy cows (de Souza et al., 2019), we observed that C16:0 supplementation consistently increased NDF digestibility ~ 5% units over the 10 weeks of treatment compared with control (Figure 1B).

## Effects of C16:0, C18:0, and C18:1 on Fatty Acid Digestibility

Our recent FA digestibility research has utilized and focused on C16:0, C18:0, and C18:1. Of particular importance, Boerman et al. (2017) fed increasing levels of a C18:0enriched supplement (93% C18:0) to midlactation 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 2A). Similarly, Rico et al. (2017) fed increasing levels of a C16:0-enriched supplement (87% C16:0) to mid-lactation dairy cows and even though a positive effect was observed on production response up to 1.5% diet DM, a decrease in total FA digestibility with increasing FA intake was observed (Figure 2B). However, considering that the range in FA intake was similar across both studies, the decrease in total

FA digestibility was more pronounced when there was increased intake/rumen outflow of C18:0 rather than C16:0. This is supported by our meta-analysis, in which a negative relationship between the total flow and digestibility of FA was observed, with the decrease in total FA digestibility driven by the digestibility of C18:0 because of the negative relationship between duodenal flow and digestibility of C18:0 (Boerman et al., 2015). The exact mechanisms for these differences in digestibility are not understood; however, potential causes include the lower solubility of C18:0 compared to C16:0, which would be more dependent of emulsification for absorption (Drackey, 2000). Additionally, results have shown that C18:1 has greater digestibility than C16:0 and C18:0 (Boerman et al., 2015). Freeman (1969) examined the amphiphilic properties of polar lipid solutes and found that C18:1 had a positive effect on the micellar solubility of C18:0. To further understand what factors influence FA digestibility, we utilized a random regression model to analyze available individual cow data from 5 studies that fed a C16:0-enriched supplement to dairy cows. We observed that total FA digestibility was negatively impacted by total FA intake, but positively influenced by the intake of C18:1 (unpublished results). This is supported by a recent study in which abomasal infusion of C18:1 increased FA digestibility without negatively affecting feed intake (Prom et al., 2018). Finally, we recently evaluated the effects of varying the ratio of dietary C16:0, C18:0, and C18:1 in basal diets containing soyhulls or whole cottonseed on FA digestibility. We observed that feeding a supplement containing C16:0 and C18:1 increased FA digestibility compared with a supplement containing C16:0, a mixture C16:0 and C18:0, and a non-fat control diet. The supplement containing a mixture C16:0 and C18:0 reduced FA digestibility compared with the other treatments (de Souza et al., 2018a). This is displayed in Figure 3 by using a Lucas

test to estimate the apparent digestibility of the supplemental FA blends. The slopes (i.e., digestibility of the supplemental FA blends) in soyhulls based diets were 0.64, 0.55 and 0.75 and in cottonseed diets were 0.70, 0.56 and 0.81 for supplements containing C16:0, a mixture C16:0 and C18:0, and a mixture of C16:0 and C18:1, respectively. This supports the concept that a combination of 16-carbon and unsaturated 18-carbon FA may improve FA digestibility, but reasons for this need to be determined.

In fresh cows, there is scarce information about the effects of supplemental FA on FA digestibility. We recently conducted a study to evaluate the effects of timing of C16:0 supplementation on nutrient digestibility of early lactation dairy cows (de Souza et al., 2019). We observed a treatment by time interaction for C16:0 supplementation during the fresh period (1 – 24 DIM); although C16:0 reduced total FA digestibility compared with control, the magnitude of difference reduced over time (Figure 4). Interestingly, we also observed an interaction between time of supplementation and C16:0 supplementation during the peak period (25-67 DIM), due to C16:0 only reducing FA digestibility in cows that received the control diet in the fresh period. This may suggest an adaptive mechanism in the intestine when C16:0 is fed long-term. In a recent study, increasing C18:1 in the FA supplement blends during early lactation increased digestibility, resulting in increased energy intake (de Souza et al., unpublished). Understanding the mechanisms responsible for this effect deserves future attention, as does the impact of other supplemental FA during early post-partum on FA digestibility and nutrient digestibility.

# Effects of C16:0, C18:0, and C18:1 on Production Responses

We have recently carried out a series of studies examining the effect of individual

saturated FA on production and metabolic responses of lactating cows. In a dose response study with mid lactation cows, feeding a C18:0enriched supplement (93% C18:0) increased DMI but had no effect on the yields of milk or milk components when compared to a non-FA supplemented control diet, which was probably associated with the decrease in FA digestibility (Figure 2A, Boerman et al., 2017). Our results, and those of others, indicate that C16:0 supplementation has the potential to increase yields of energy corrected milk (ECM) 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 (Rico et al., 2014; de Souza et al., 2018a). We recently utilized a random regression model to analyze available individual cow data from 10 studies whereby C16:0-enriched supplements were fed to post peak dairy cows (de Souza et al., 2016). 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 ECM with increasing intake of C16:0.

When we compared combinations of C16:0, C18:0, and C18:1 in FA supplements, a supplement containing more C16:0 increased energy partitioning toward milk due to the greater milk fat yield response compared with the other treatments (de Souza et al., 2018a). In contrast, a FA supplement containing C16:0 and C18:1 increased energy allocated to body reserves compared with other treatments. The FA supplement containing a combination of C16:0 and C18:0 reduced nutrient digestibility, which most likely explains the lower production responses observed compared with the other treatments. Interestingly, in a follow up study, we compared different ratios of C16:0 and C18:1 in FA supplements fed to post-peak cows and observed that supplements with more C16:0 favored energy partitioning to milk in cows producing less than 45 kg/day, while

supplements with more C18:1 favored energy partitioning to milk in cows producing greater than 60 kg/day (de Souza and Lock, 2017). Also, regardless of production level, supplements with more C18:1 increased BW change. This may suggest that C16:0 and C18:1 are able to alter energy partitioning between the mammary gland and adipose tissue, which may allow for different FA supplements to be fed in specific situations according to the metabolic priority and needs of dairy cows. Further research is needed to confirm these results in cows at different stages of lactation or other physiological conditions.

In early lactation cows, Beam and Butler (1998) fed a saturated FA supplement (~ 40% C16:0 and 40% C18:0) and observed that FA supplementation decreased DMI and did not affect yields of milk and ECM in the first 4 weeks after calving. Piantoni et al. (2015b) fed a similar saturated FA supplement (~ 40% C16:0 and 40% C18:0) and observed that FA supplementation during the immediate postpartum (1 to 29 DIM) favored energy partitioning to body reserves rather than milk yield, especially in the lower forage diet. The high forage diet with supplemental FA increased DMI and tended to decrease body condition score (BCS) loss compared with the same diet without FA supplementation. Also, regardless of forage level, feeding supplemental FA increased DMI, decreased BCS loss, but tended to decrease milk yield. When cows were fed a common diet during the carryover period, the low forage diet with FA supplementation fed during the immediate postpartum continued to decrease milk yield and maintained higher BCS compared with the other treatments. On the other hand, Weiss and Pinos-Rodriguez (2009) fed a similar saturated FA supplement (~ 40% C16:0 and 40% C18:0) to early-lactation cows (21 to 126 DIM) and observed that when high-forage diets were supplemented with FA, the increased NEL intake went toward body energy reserves as

measured by higher BCS with no change in milk yield. However, when low-forage diets were supplemented with FA, milk yield increased (2.6 kg/day) with no change in BCS.

We evaluated the effects of timing of C16:0 supplementation on performance of early lactation dairy cows (de Souza and Lock, 2019). During the fresh period (1 to 24 DIM), we did not observe treatment differences for DMI or milk yield (Figure 5A), but compared with control, C16:0 increased the yield of ECM by 4.70 kg/day consistently over time (Figure 5B). However, C16:0 reduced BW by 21 kg (Figure 6), and BCS by 0.09 units and tended to increase BW loss by 0.76 kg/day compared with CON. Feeding C16:0 during the peak period (25 to 67 DIM) increased the yield of milk by 3.45 kg/ day, ECM yield by 4.60 kg/day (Figure 5), and tended to reduce BW by 10 kg compared with control (Figure 6).

In a recent study, a non-FA supplemented control diet was compared with diets supplemented at 1.5% DM with FA supplements differing in the ratio of C16:0 and C18:1 (de Souza et al., 2018b). FA treatment diets were: 80:10 (80% C16:0 + 10% C18:1); 70:20 (70% C16:0 + 20% C18:1); and 60:30 (60% C16:0 + 30% C18:1). From days 25 to 60 postpartum, all cows were offered a common diet to evaluate carryover effects. During the fresh period, FA-supplemented diets increased milk yield, ECM, and milk fat yield (Figure 7). Increasing C18:1 in FA treatments decreased plasma NEFA and BW loss and tended to increase DMI and plasma insulin (Figure 7). Increasing C18:1 in FA treatments did not affect milk yield, ECM, and the yields of milk fat and protein. During the carryover period, cows that received FAsupplemented diets during the fresh period increased ECM and milk fat yields compared with the control treatment.

Interestingly, Greco et al. (2015) observed that decreasing the ratio of omega-6 to omega-3 FA in the diet of lactating dairy cows while maintaining similar dietary concentrations of total FA improved productive performance in early lactation. A dietary omega-6 to omega-3 ratio of approximately 4:1 increased DMI and production of milk and milk components compared with a 6:1 ratio. Approximately 1.3 kg of milk response could not be accounted for by differences in nutrient intake, which suggests that reducing the dietary FA ratio from 6:1 to 4:1 can influence nutrient partitioning to favor an increased proportion of the total net energy consumed allocated to milk synthesis. Further studies focusing on altering ratio of dietary FA are warrant, especially in early lactation cows.

# **Effects of Supplemental Fatty Acids on Reproduction**

A recent meta-analysis of 17 studies reported a 27% increase in pregnancy rate in the first postpartum artificial insemination (AI) when dairy cows were fed fat supplements during the transition period (Rodney et al., 2015). In addition, the interval from calving to pregnancy was reduced. The inclusion of the very long chain omega-3 FA eicosapentaenoic acid (EPA) and docosahexaenoic acid (DHA) in the form of fish meal, fish oil, or algae in the diet has been shown to improve either first-service or over- all pregnancy in 6 studies (Santos and Staples, 2017). A study conducted at the University of Florida (Silvestre et al., 2011) demonstrated that supplementation with Ca salts (1.5% of dietary DM) enriched in fish oil-derived FA starting at 30 DIM improved pregnancy rate/AI compared with Ca salts of palm FA (52.8 vs. 45.5%). Additionally, pregnancy loss between 32 and 60 days after AI was reduced by feeding Ca salts containing EPA and DHA (6.1 vs. 11.8%). Recently, Sinedino et al. (2017) observed that feeding 100 g/day

of an algae product containing 10% of DM as DHA starting in the third week postpartum increased pregnancy rate by 39% and reduced days to pregnancy by 22 days (102 vs. 124 days). Therefore, polyunsaturated long-chain FA including omega-6 and omega-3 seem to be more effective at improving pregnancy in dairy cows than those containing mainly C16:0 and C18:1. Furthermore, a meta-analysis indicated that the probability of pregnancy increased by 26% and the days from calving to pregnancy decreased by 34 days when trans-10, cis-12 conjugated linoleic acid was fed as a Ca-salt product across 5 studies involving 221 early lactation dairy cows (de Veth et al., 2009). Feeding long-chain FA might improve reproduction in dairy cattle through several potential mechanisms, including reducing negative energy balance, changes in follicle development and improvements in oocyte quality, improved early embryo development, and reduced pregnancy loss. Since individual FA have a direct effect on several metabolic processes, research should focus on determining "ideal" combinations of FA for cows under specific physiological conditions and for specific purposes.

#### **Conclusions**

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 improve reproduction performance, great variation has been reported in production performance for different FA supplements, and indeed, the same supplement across different diets and studies. Results are contradictory about the benefits of FA supplementation to early lactation dairy cows. We propose that this is a result of differences in FA profile of supplements used and the time at which FA supplementation starts. However,

our recent results suggest the use of specific supplemental FA and FA blends in the fresh period should be considered; however, 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 sources and 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, body condition, and their effects on immune and reproductive functions. The extent of these simultaneous changes along with the goal of the nutritional strategy employed will ultimately determine the overall effect of the FA supplementation, and the associated decision regarding their inclusion in diets for lactating dairy cows.

#### References

Allen, M.S., and P. Piantoni. 2013. Metabolic control of feed intake: implications for metabolic disease of fresh cows. Veterinary Clinics of North America: Food Animal Practice. 29:279-297.

Beam, S.W., and W.R. Butler. 1998. Energy balance, metabolic hormones, and early postpartum follicular development in dairy cows fed prilled lipid. J. Dairy Sci. 81:121–131.

Boerman, J.P., J. de Souza, and A.L. Lock 2017. Milk production and nutrient digestibility responses to increasing levels of stearic acid supplementation of dairy cows. J. Dairy Sci. 100: 2729:2738.

Boerman, J.P., J.L. Firkins, N.R. St-Pierre, and A.L. Lock. 2015. Intestinal digestibility of long-chain fatty acids in lactating dairy cows: A meta-analysis and meta-regression. J. Dairy Sci. 98:8889–8903.

de Souza, J., and A.L. Lock. 2017. Altering the ratio of dietary C16:0 and cis-9 C18:1 interacts with production level in dairy cows: Effects on production responses and energy partitioning. J. Dairy Sci. 100 (E-Suppl. 1):221.

de Souza, J., and A.L. Lock. 2019. Effects of timing of palmitic acid supplementation on production responses of early-lactation dairy cows. J. Dairy Sci. 102:260–273.

de Souza, J., C.L. Preseault, and A.L. Lock. 2018a. Altering the ratio of dietary palmitic, stearic, and oleic acids in diets with or without whole cottonseed affects nutrient digestibility, energy partitioning, and production responses of dairy cows. J. Dairy Sci. 101:172–185.

de Souza, J., C.M. Prom, and A.L. Lock. 2018b. Altering the ratio of dietary palmitic and oleic acids impacts production and metabolic responses during the immediate postpartum and carryover period in dairy cows. J. Dairy Sci. 101 (Suppl. 2): 160.

de Souza, J., C. Strieder-Barboza, G.A. Contreras, and A.L. Lock. 2019. Effects of timing of palmitic acid supplementation during early lactation on nutrient digestibility, energy balance, and metabolism of dairy cows. J. Dairy Sci. 102:274–287.

de Souza, J., R.J. Tempelman, M.S. Allen, and A.L. Lock. 2016. Production response, nutrient digestibility, and energy partitioning of postpeak dairy cows when palmitic acid-enriched supplements are included in diets: A meta-analysis and meta-regression. J. Dairy Sci. 99 (E-Suppl. 1):622.

de Veth, M.J., D.E. Bauman, W. Koch, G.E. Mann, A.M. Pfeiffer, and W.R. Butler. 2009. Efficacy of conjugated linoleic acid for improving reproduction: A multi-study analysis in early-lactation dairy cows. J. Dairy Sci. 92:2662–2669.

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.

Freeman, C.P. 1969. Properties of FA 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.

Greco, L.F., J.T.N. Neto, A. Pedrico, R.A. Ferrazza, F.S. Lima, R.S. Bisinotto, N. Martinez, M. Garcia, E.S. Ribeiro, G.C. Gomes, J.H. Shin, M.A. Ballou, W.W. Thatcher, C.R. Staples, and J.E.P. Santos. 2015. Effects of altering the ratio of dietary n-6 to n-3 fatty acids on performance and inflammatory responses to a lipopolysaccharide challenge in lactating Holstein cows. J. Dairy Sci. 98:602–617.

Kuhla, B., C. C. Metges, and H. M. Hammon. 2016. Endogenous and dietary lipids influencing feed intake and energy metabolism of periparturient dairy cows. Dom. Anim. Endoc. 56:S2–S10.

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

Piantoni, P., A.L. Lock, and M.S. Allen. 2015b. Saturated fat supplementation interacts with dietary forage neutral detergent fiber content during the immediate postpartum and carryover periods. Production responses and digestibility of nutrients. J. Dairy Sci. 98:3309–3322.

Prom, C.M., J. Newbold, and A.L. Lock. 2018. Impact of abomasal infusion of oleic acid on fatty acid digestibility and milk production of dairy cows. J. Dairy Sci. 101 (Suppl. 2): 159.

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.

Rico, J. E., J. de Souza, M. S. Allen, and A. L. Lock. 2017. Nutrient digestibility and milk production responses to increasing levels of palmitic acid supplementation vary in cows receiving diets with or without whole cottonseed. J. Anim. Sci. 95: 434 – 446.

Rodney, R.M., P. Celi, W. Scott, K. Breinhild, and I.J. Lean. 2015. Effects of dietary fat on fertility of dairy cattle: A meta-analysis and meta-regression. J. Dairy Sci. 98:5601–5620.

Santos, J.E.P., and C.R. Staples. 2017. Feeding the herd for maximum fertility. In: Large Dairy Herd Management, 3rd ed, American Dairy Science Association.

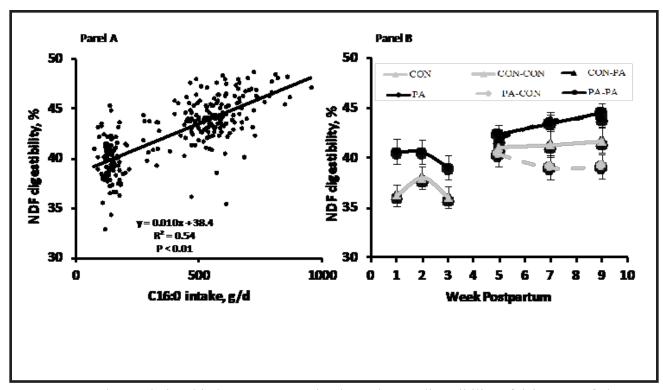
Silvestre, F.T., T.S.M. Carvalho, N. Francisco, J.E.P. Santos, C.R. Staples, T. Jenkins, and W.W. Thatcher. 2011. Effects of differential supplementation of fatty acids during the peripartum and breeding periods of Holstein cows: I. Uterine and metabolic responses, reproduction, and lactation. J. Dairy Sci. 94:189–204.

Sinedino, L.D. P., P.M. Honda, L.R.L. Souza, A.L. Lock, M.P. Boland, C.R. Staples, W.W. Thatcher, and J.E.P. Santos. 2017. Effects of supplementation with docosahexaenoic acid on reproduction of dairy cows. Reproduction <a href="https://doi.org/10.1530/REP-16-0642">https://doi.org/10.1530/REP-16-0642</a>.

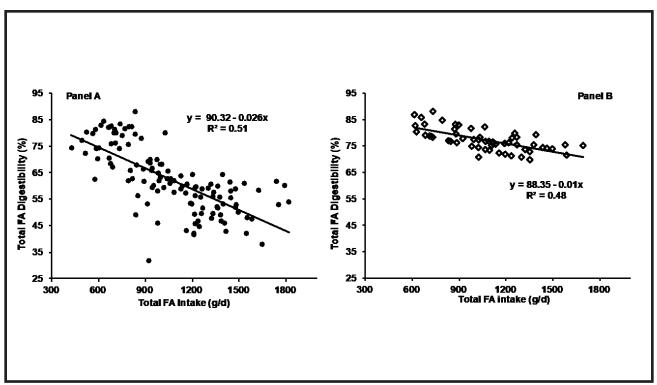


Weiss, W.P., and J.M. Pinos-Rodríguez. 2009. Production responses of dairy cows when fed supplemental fat in low- and high-forage diets. J. Dairy Sci. 92:6144–6155.

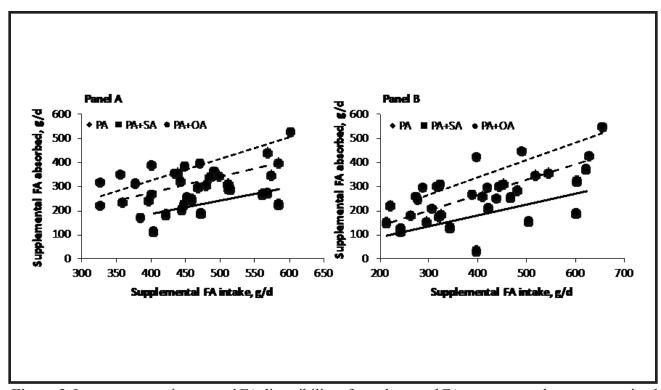
Weld, K.A. and L.E. Armentano. 2017. The effects of adding fat to diets of lactating dairy cows on total-tract neutral detergent fiber digestibility: A meta-analysis J. Dairy Sci. 100: 1766-1779.



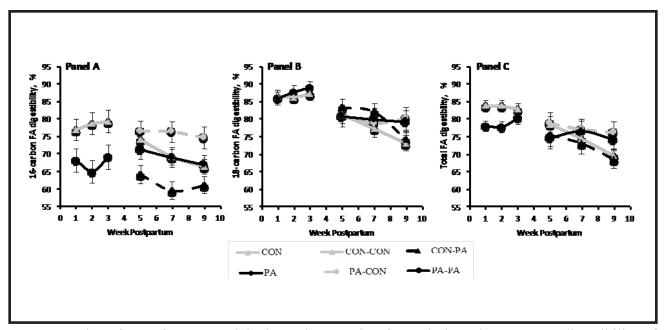
**Figure 1.** Panel A: Relationship between C16:0 intake and NDF digestibility of dairy cows fed C16:0-enriched FA supplements. Panel B: The effects of C16:0-enriched supplementation in early lactation cows on NDF digestibility. Results in Panel A represent a combined data set evaluated using a random regression model from 6 studies feeding C16:0-enriched supplements on NDF digestibility of post-peak cows (de Souza et al., 2016). Results in Panel B utilized 52 early-lactation cows receiving the following diets: no supplemental fat (CON) or a C16:0 supplemented diet (PA) that was fed either from calving (1 to 24 DIM; fresh period) or from 25 to 67 DIM (peak period). From de Souza et al. (2019).



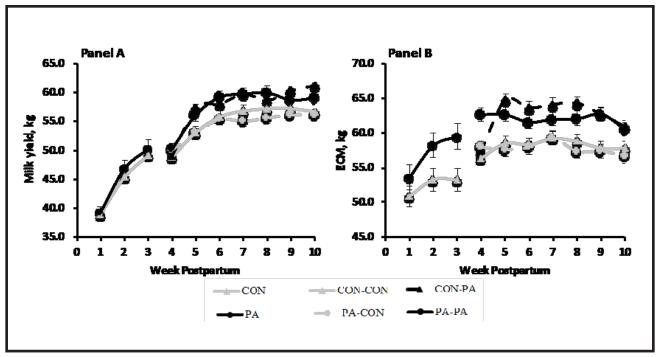
**Figure 2.** Relationship between total FA intake and apparent total-tract FA digestibility of dairy cows supplemented with either a C18:0-enriched supplement (Panel A) or a C16:0-enriched supplement (Panel B). Results in Panel A utilized 32 mid-lactation cows receiving diets with increasing levels (0 to 2.3% dry matter) of a C18:0-enriched supplement (93% C18:0) in a 4 X 4 Latin square design with 21-d periods (Boerman et al., 2017). 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 (Rico et al., 2017).



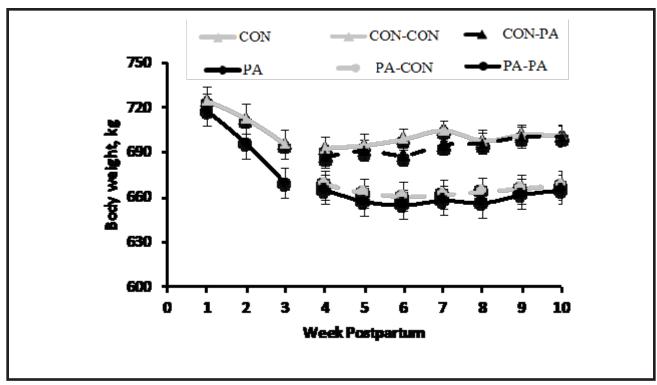
**Figure 3.** Lucas test to estimate total FA digestibility of supplemental FA treatments when cows received either a soyhulls basal diet (Panel A) or a cottonseed basal diet (Panel B) PA long-dashed line (1.5% of FA supplement blend to provide  $\sim 80\%$  of C16:0); PA+SA solid line (1.5% of FA supplement blend to provide  $\sim 40\%$  of C16:0 + 40% of C18:0); and PA+OA short-dashed line (1.5% of FA supplement blend to provide  $\sim 45\%$  of C16:0 + 35% of C18:1). Digestibility of supplemental FA was estimated by regressing intake of supplemental FA on intake of digestible supplemental FA. The mean intakes of FA and digestible FA when cows were fed the control diet were subtracted from the actual intakes of total FA and digestible FA for each observation. From de Souza et al. (2018a).



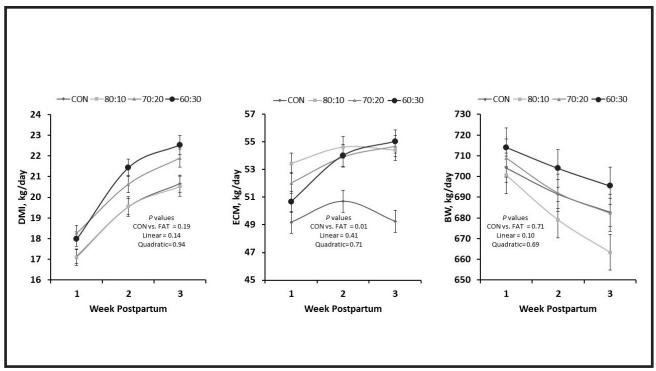
**Figure 4.** The effects of C16:0-enriched supplementation for early lactation cows on digestibility of 16-carbon (Panel A), 18-carbon (Panel B), and total FA (Panel C). Results utilized 52 early-lactation cows receiving the following diets: no supplemental fat (CON) or a C16:0 supplemented diet (PA) that was fed either from calving (1 to 24 DIM; fresh period FR) or from 25 to 67 DIM (peak period). From de Souza et al. (2019).



**Figure 5.** The effects of C16:0-enriched supplementation in early lactation cows on the yield of milk (Panel A) and ECM (Panel B). Results from 52 early-lactation cows receiving the following diets: no supplemental fat (CON) or a C16:0 supplemented diet (PA) that was fed either from calving (1 to 24 DIM; fresh period FR) or from 25 to 67 DIM (peak period). From de Souza and Lock (2019).



**Figure 6.** The effects of C16:0-enriched supplementation in early lactation cows on body weight. Results from 52 early-lactation cows receiving the following diets: no supplemental fat (CON) or a C16:0 supplemented diet (PA) that was fed either from calving (1 to 24 DIM; fresh period) or from 25 to 67 DIM (peak period). From de Souza and Lock (2019).



**Figure 7.** The effects of altering the C16:0 to C18:1 ratio of supplemented fats in early lactation cows on DMI, milk yield, and BW. Results from 52 early-lactation cows receiving the following diets: no supplemental fat (CON) and diets supplemented at 1.5% DM with FA supplements differing in the ratio of palmitic (C16:0) and oleic (C18:1) acids. FA treatment diets were: 80:10 (80% C16:0+10% C18:1); 70:20 (70% C16:0+20% C18:1); and 60:30 (60% C16:0+30% C18:1). From de Souza et al. (2018b).