

# Using Fat Supplementation to Improve the Chances of Pregnancy of Lactating Dairy Cows

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## Historical Decreases in Reproductive Efficiency

The proportion of lactating dairy cows on commercial farms that become pregnant at the first insemination has decreased over the last 25 to 30 years. Data from New York herds indicate that the first service conception rates decreased from ~55% in 1975 to just under 40% in 2001 (Butler, 2005). The number of services per conception increased from 1.62 in 1970 to 2.91 in 1999 in Kentucky herds (Silva, 1998). The DHIA records from Raleigh, NC show that the 143 herds on DHIA for 29 consecutive years (1970 to 1999) experienced a decrease in services per conception from ~1.75 to just under 3.0 (Lucy, 2001). Conception rates measured for cows managed under controlled experimental conditions as reported in scientific journals also have decreased. Rates dropped from ~55% to ~45% (breeding at spontaneous estrus) to ~35% (timed AI) over a 50-year period (Lucy, 2001).

The dollar value assigned to pregnancy for dairy cows varies because it is dependent upon several factors, such as how many days she has been milking, her lactation number, her milk yield, the replacement costs of a pregnant heifer, milk price, etc. A modeling program developed at the University of Florida was used to predict pregnancy value. Some key input values were a milk price of \$14.09 per 100 pounds of milk, 305-d milk production of 23,144 lb for young animals and 25,994 lb for third lactation cows, and a replacement heifer cost of \$1600 per head. The value of a new pregnancy at about 100 days in milk was calculated to be ~\$200 for a milking heifer and ~\$300 for a cow in her second lactation (de Vries, 2006). Even when a cow conceives, the pregnancy does not go to term about 50% of the time. If an average producing cow in the herd conceived at 61 days in milk, but was declared open 30 days later, the calculated loss ranged from \$110 for heifers to \$336 for cows in their third lactation (de Vries, 2006). Efforts to reduce this loss are certainly justified.

Many reasons for these declines in reproductive efficiency have been offered, including an increase in postpartum disease (ketosis, mastitis, retained fetal membranes, cystic ovaries, fatty liver, etc.), an increase in herd size resulting in increased management challenges, an increase in the proportion of milking heifers in the herd which cycle later, an increase in genetic inbreeding, and an increase in milk production (Lucy, 2001). Average milk production per lactation has increased by 57% from ~12,000 to ~19,000 pounds per cow in the last 25 years (Eastridge, 2006). However, amount of milk production has not been an accurate predictor of the chance for pregnancy. For example, higher-producing cows that ate very well cycled sooner after calving than lower-producing cows that ate poorly (Staples et al., 1990). Those poor eaters lost more

body weight in the first 2 weeks postpartum and had not cycled by 9 weeks postpartum and fewer became pregnant. Extent of negative energy balance (energy output in milk plus body maintenance minus energy intake from the diet) might be a more important factor influencing pregnancy. It is not known if the modern-day dairy cow is in a more negative energy state than dairy cows from 30 years ago. Nevertheless, the lactating cow is more difficult to get pregnant than the nonlactating cow. In conclusion, several suspects have been identified that may be contributing to this lowered fertility, what about some management steps to help promote reproductive efficiency?

## **Fat Supplementation and Reproduction Introduction**

The influence of nutrient intake on reproductive performance is a growing field of study, including the effect of feeding supplemental fat. Supplementing fat (the most energy dense nutrient) in small amounts seems reasonable because milk production is such an energy-demanding process. Fat supplementation in moderate amounts often stimulates milk production and has improved reproductive efficiency. However its impact on reproduction has not been consistent. Questions such as which fat sources can be most effective and how do these fats work have not yet been adequately answered. The purpose of this paper is to review some of the effects of fat supplementation on reproductive tissues and pregnancy.

### **Fats Defined**

Many different types of supplemental fat have been fed to lactating cows. Some fat sources fed are listed in Table 1. Each fat source is composed of a different mix of individual fatty acids. Rendered fats include animal tallow and yellow grease (recycled restaurant grease) and are composed mainly of oleic acid (~43%). Granular fats are dry fats prepared commercially and are composed mainly of palmitic acid (36-50%). Examples include Energy Booster 100, EnerG-II, and Megalac-R. A variety of vegetable oils can be fed as free oil or in the seed form. The oil seeds contain from 18% oil (such as soybeans) to 40% oil (such as flaxseed). The selection of a vegetable oil will bring with it particular fatty acids. Canola oil is high in oleic acid. Cottonseed, safflower, sunflower, and soybean oils are high in linoleic acid. Flaxseed is high in linolenic acid. Linoleic acid and linolenic acid are essential fatty acids for the cow because neither her body nor her ruminal microorganisms can synthesize them. Fresh temperate grasses contain 1 to 3% fatty acids of which 55 to 65% is linolenic acid (Chilliard et al., 2001). Corn silage lipid contains much more linoleic acid (49%) than linolenic acid (4%) due to the presence of corn grain (Petit et al., 2004). Both linoleic and linolenic acid in forages can decrease during storage. As we have moved our dairy cows from pastures to barns and fed them stored forage, their intake of linolenic acid and possibly linoleic acid has likely decreased. The whole oil seed is frequently fed rather than the oil alone. Fish oil is unique that it contains eicosapentaenoic acid (EPA) and docosahexaenoic acid (DHA), fatty acids found in fish tissue due to their consumption of marine plants.

Table 1. Major fatty acid composition of select dietary fat sources.

Fat source	Fatty acid						
	C14:0 Myristic	C16:0 Palmitic	C16:1 Palmit- oleic	C18:0 Stearic	C18:1 Oleic	C18:2 Linoleic	C18:3 Linolenic
Tallow	3	25	3	18	43	3.8	<1
Yellow grease	2	21	4	11	44	14	<1
Energy Booster 100 <sup>1</sup>	3	40	1	41	10	2	<1
Megalac; EnerG-II <sup>1</sup>	1	50	<1	4	36	8	<1
Megalac-R <sup>1</sup>	1	36	<1	4	26	29	3
Canola oil	<1	4	<1	2	63	19	9
Cottonseed oil	1	23	1	3	18	54	1
Flaxseed oil	<1	5	<	3	20	16	55
Rapeseed oil	<1	5	<1	2	54	22	11
Safflower oil	<1	7	<1	2	12	78	<1
Soybean oil	<1	11	<1	4	23	54	8
Sunflower oil	<1	7	<1	5	19	68	1
Menhaden fish oil <sup>2</sup>	7	16	8	3	12	1	2

<sup>1</sup>Commercial preparations considered partially inert in the rumen.  
<sup>2</sup>Also contains 14% C20:5 and 9% C22:6.

The short-hand notation for identifying fatty acids is to give the number of carbons and double bonds in the molecule. Fats that have double bonds are classified as unsaturated fats. For example, a designation of 18:2 indicates a fatty acid of 18 carbons long having 2 double bonds. The term “omega” refers to the location of the double bond in the carbon chain. An omega-6 fatty acid has its first double bond located between the 6<sup>th</sup> and 7<sup>th</sup> carbon counting from the methyl end of the chain. Likewise an omega-3 fatty acid has its first double bond located between the 3<sup>rd</sup> and 4<sup>th</sup> carbon counting from the methyl end of the carbon chain. Linoleic acid, abbreviated C18:2, is an omega-6 fatty acid. Linolenic acid, abbreviated C18:3, is an omega-3 fatty acid. Two additional omega-3 fatty acids are EPA (C20:5) and DHA (C22:6); but these are not considered essential for the cow because they can be synthesized from the omega-3 fatty acid, linolenic acid. Nevertheless EPA and DHA can play important roles in supporting good animal performance.

### **Dietary Fats Are Modified in the Rumen by Bacteria**

The ruminal microbes will convert unsaturated fats to saturated fats by replacing the double bonds with single bonds between the carbons (called biohydrogenation). Some scientists have speculated that this act of biohydrogenation by bacteria is an attempt to protect themselves, as unsaturated fats can be toxic to bacteria, primarily the bacteria that digest fiber. The majority of the consumed unsaturated essential fatty acids, linoleic (C18:2) and linolenic (C18:3) acids, are converted by the bacteria to stearic acid (C18:0). During the process of biohydrogenation of unsaturated fats in the rumen, several intermediate forms of fatty acids, called trans fatty acids, also are formed. Some of the trans fatty acids, such as the trans-10, cis-12 conjugated linoleic acid (CLA) and the trans-10 C18:1, can influence the cow’s metabolism, including depressing milk fat synthesis. This intervention by ruminal bacteria to change essential fatty acids in the diet to other fatty acids has made the study of dietary fat effects on reproduction quite challenging.

## Fat Supplementation and Conception Rates

According to the scientific literature, a variety of fat supplements have benefited conception rates of lactating dairy cows (Table 2). The conception rates are sometimes reported for first insemination or for cumulated inseminations. Feedstuffs stimulatory to conception included calcium salts of palm oil distillate, tallow, Energy Booster (prilled tallow), MegaPro Gold (which is a calcium salt of palm oil plus rapeseed meal and whey permeate) fed to grazing cows, flaxseed (formaldehyde-treated or rolled), calcium salt of a mixture of soy oil and monounsaturated trans fatty acids), CLA, Megalac-R, and fish meal. The average improvement in conception rate was 21 percentage units. This is not to imply that the feeding of one of these feedstuffs to cows on your farm will increase herd conception rate by 21 percentage units. The margin of increase in conception rate due to fat feeding needed to be very great in these trials involving a few number of cows in order for the fat supplement to be detected as having a significant effect. If a fat supplement is to be beneficial on a dairy farm, it is more likely that the benefit will be less than 10 percentage units. In 3 of the 4 studies in which at least 250 cows participated in the study, the margin of increase due to fat supplementation ranged from 5 to 9 percentage units (Table 2). It is somewhat surprising that fat supplementation improved conception in experiments using between 30 and 132 cows. This may be partially due to the tighter management of cows used in experiments compared to that used on commercial farms. Normally about 300 cows per treatment are required to have a high chance of detecting a 10% increase in conception rate due to a treatment. Certainly other studies have been published in which fat-supplementation did not improve pregnancy rate. Staples et al. (1998) lists some of those studies. On the beef cow side, Funston (2004) reviewed several studies and concluded that the effects of fat supplementation on reproductive function were inconsistent but that benefits may depend upon the quality of the pastures grazed.

In these studies, the fat-supplemented diet (last column in Table 2) was compared to a control diet that contained no supplemental fat in some studies, whereas in other studies the control diet contained another fat source. In head-to-head comparisons of fat sources, flaxseeds were more effective than rolled sunflower seeds (Canada, 2006). Based on this, we might be tempted to conclude that the omega-3 fatty acid (linolenic acid) found in flaxseed is more effective than the omega-6 fatty acid (linoleic acid) found in sunflower seeds. However the linoleic acid in sunflower seeds is extensively biohydrogenated by the ruminal bacteria so that little is absorbed by the cow for reproductive use. This is based upon information that the linoleic acid in milk was not changed when cows were fed sunflower seeds (Petit et al., 2004). In two other head-to-head comparisons of fat supplements, cows fed calcium salts of palm oil distillate did not conceive as well as those fed either flaxseed (Canada, 2001) or those fed a calcium salt mixture of soybean oil and monounsaturated trans fatty acids (California, 2004) (Table 2). Therefore fats containing more linolenic acid (flaxseed) or linoleic acid (soybean oil) may be more effective than fats containing palmitic and oleic acid. However, as shown in Table 2, cows fed fats containing mainly palmitic and oleic acids such as tallow (Nebraska, 1996) and Energy Booster (New York, 2003) benefited cows reproductively. Possibly the trans fatty acids synthesized in the rumen by bacteria play a positive role for improving conception. Some of these trans fats can now be manufactured and fed to cows. The positive benefit to conception observed in the California study (34 vs. 26%) may have been due to the supplementation of the trans fat. Supplementing with trans fats (CLA) also appeared to benefit lactating cows in New York

compared to those fed a calcium salt of palm oil distillate (81 vs. 44%) (Table 2). Increased synthesis of trans fats in the rumen is favored when cows are fed supplemental unsaturated fatty acids with high grain diets.

Table 2. Studies Reporting Improved Conception Rates (first service or cumulative services) of Lactating Dairy Cows Fed Supplemental Fatty Acids ( $P < 0.10$ ). Unless otherwise indicated with a footnote, the control diet did not contain a fat supplement.

Reference	Fat source	Number of cows in trial	Control treatment	Fat treatment
			----- % -----	
Penn & Israel, 1990	2% Ca-Palm oil	253	43	59 <sup>1</sup>
Israel, 1991	2.6% Ca-Palm Oil	99	62	82
Wisconsin, 1995	1 lb/d Ca-Palm oil	443	93	98
Florida, 1998	2.2% Ca-Palm oil	43	52	86
Nebraska, 1996	3% Tallow	68	44	62
New York, 2003	1.7% Energy Booster	81	58 <sup>2</sup>	86
United Kingdom, 2001	17% Flaxseed	30	50 <sup>3</sup>	87
Canada, 2006	9% Flaxseed	74	73 <sup>4</sup>	90
Ireland, 2003	3.3 lb/d MegaPro Gold	129	35	54
California, 2004	1.5% Soy + Trans C18:1	397	26 <sup>3</sup>	34 <sup>1</sup>
Florida, 2004	2% Megalac-R	42	27	58 <sup>1</sup>
New York, 2005	0.3 lb/d Ca-CLA	32	44 <sup>3</sup>	81
UK, 1989	7.3% Fish meal	132	52	72
Ireland, 1990	1.8 lb/d fish meal	80	44	64
Oregon, 1994	3.5% fish meal	62	68	89
Florida, 1997	2.8% fish meal	300	32	41
Average			50.2	71.4

<sup>1</sup> First insemination.

<sup>2</sup> Control diet contained equal energy diet to fat-supplemented diet. Fat was fed prepartum only.

<sup>3</sup> Control diet contained Ca salt of palm oil distillate.

<sup>4</sup> Control diet contained rolled sunflower seeds.

Although the main nutrient in fish meal is protein and not fat, it is included here because there is growing evidence that the oils unique to fish may play a very important role in establishing pregnancy. The inclusion of fish meal in the diet (2.7 to 7.3% of dietary DM) also has improved either first service or overall pregnancy rate in four studies. In some of these studies, fish meal partially replaced soybean meal resulting in a reduction of an excessive intake of ruminally degradable protein. Therefore, the improved conception rates may have been due to the elimination of the negative effect of excessive intake of ruminally degradable protein on conception. However, in a field study in which the concentration of ruminally undegradable protein was kept constant between dietary treatments, cows fed fish meal had a better conception rate (Burke et al., 1997) suggesting that the positive response was due to something other than a reduction in intake of ruminally degradable protein. The unique omega-3 polyunsaturated fatty acids in fish (EPA and DHA) may have been responsible for the improvement in fertility, hence their inclusion in the current discussion.

Oil seeds have not been well evaluated for their ability to improve conception. Those seeds that can deliver the key fatty acids past the rumen may be good candidates for the diet. Although the oil in many oil seeds contains more than 50% linoleic acid (Table 1), the delivery of linoleic acid past the rumen to the small intestine is not the same for all oil seeds. If we use an increase in the linoleic acid concentration of milk as an indicator that an oil seed can deliver linoleic acid to the tissues, then soybeans appear to be most effective and cottonseeds seem to be ineffective (Table 4). Sunflower seeds and safflower seeds also can increase the linoleic acid of milk fat, but not quite as effectively as that of soybeans. The processing of whole seeds also can influence their ability to deliver unsaturated fat past the rumen. Roasting of soybeans and rolling of sunflowers seemed to increase delivery of linoleic acid. Regarding linolenic acid, whole flaxseeds fed at about 10% of the diet can deliver some of its omega-3 fatty acid to the tissues. Grinding the flaxseed may deliver even more linolenic acid to the tissues (Table 3). In the United Kingdom, a process has been developed in which cracked linseeds or soybeans are processed with steam in order to create Maillard products, which help to protect the seed's unsaturated fatty acids from ruminal microbes (Robinson et al., 2002). Obviously, more research needs to be done to better identify the most effective fat sources, whether from seeds, oils, or calcium salts.

Table 3. Effect of Feeding Various Oilseeds on the Essential Fatty Acid Concentration of Milk Fat From Dairy Cows.<sup>1</sup>

Reference	Seed type	Diet	
		Control	+ Oil Seed
		% of milk fatty acids	
C18:2			
Dhiman et al., 1995	0% vs. 16% soybeans	3.2%	6.2%*
Holter et al., 1992	0% vs. 15% whole cottonseeds	4.0%	4.2%
Markus et al., 1996	0% vs. 7.1% whole sunflower seeds	2.3%	2.8%*
Petit et al., 2004	0% or 9.6% whole sunflower seeds	3.2%	3.8%
Stegeman et al., 1992	0% or 10% rolled sunflower seeds	2.2%	3.3%*
Tice et al., 1994	19.7% raw vs. roasted whole soybeans	5.5%	6.7%*
Stegeman et al., 1992	0% or 10% rolled safflower seeds	2.2%	3.1%*
C18:3			

Petit et al., 2004	0 vs.9.7% whole flaxseed	0.6%	1.1%*
Gonthier et al., 2005	0% vs. 12.5% ground flaxseed	0.4%	1.3%*

\* Values under the oilseed column having an asterisk were significantly different from the control values.

### Amount of Fat to Feed and Economic Assessment

A frequent asked question is “How much fat or of a specific fatty acid should be fed in order to improve reproduction?” In the studies listed in Table 2, the fat sources were fed at a minimum of 1.5% of dietary dry matter. We know that feeding these amounts were effective. We do not know if feeding a smaller amount of fat would be effective as well. People are interested in feeding a smaller amount of fat for various reasons including keeping feed costs down and minimizing the potential negative effects of supplemental fats on the cow’s bacteria in her rumen. These negative effects can include reduced fiber digestion and reduced fat and protein concentrations in the milk. Generally speaking, fat supplementation at 1.5% of the diet is usually safe in terms of cow performance with the exception of fish oil. Feeding fish oil at more than 1% of dietary dry matter will usually reduce feed intake and/or milk fat and protein concentration. If the fat concentration of the base diet without a fat supplement is 3 to 4%, then increasing it to 4.5 to 5.5% by fat supplementation should not be a problem if the dietary fiber is sufficient and effective. Certainly diets containing higher fat ingredients like distillers grains, hominy, or whole cottonseeds need to be watched closely so that the total fat content stays below 6%.

It is certainly possible that feeding supplemental fat at a lower rate such as 0.25 or 0.5 pounds per day could be effective. The key fatty acids (whether it is linoleic, linolenic, trans fatty acids, EPA, DHA, or something else) that do reach the small intestine of the cow are absorbed into the blood stream and deposited into tissues including her reproductive tissues. Some of these can accumulate over time. In a Florida study, the concentration of EPA increased in the liver fat from approximately 0.05 to 0.5 to 0.9% in liver samples collected at 2, 14, and 28 days in milk from cows fed linseed oil starting 5 weeks prepartum. A small but steady supply of these key fatty acids streaming to the tissues will allow the tissues to accumulate the fatty acids and have them ready at the proper time for reproductive purposes. Therefore, even a smaller fat feeding rate than the 1.5% as used by one of the published experiments in Table 2 could prove beneficial.

The economic assessment of fat supplementation on herd performance is not a straight-forward calculation. It is based upon many factors including the base conception rate of a herd, improvement in conception rate and/or milk yield, milk price, replacement costs, etc. Based upon some preliminary calculations using the breeding and replacement model described by De Vries (2006), the economic benefit of fat supplementation to lactating dairy cows was evaluated. If conception rate was improved from 17 to 20% and milk production (\$14/cwt) was improved by 2 pounds per day through supplementation of fat, an additional \$100 per cow would be realized. If the fat supplement was fed at 1.5% of dietary dry matter, intake of fat would be about 0.7-0.8 pounds per day. The breakeven price in the diet to realize this benefit would be \$0.38 per cow per day over 300 days (\$100/300 days plus the \$0.05/day saving on replacing corn (\$120/ton) with fat). To have a 2:1 benefit:cost ratio, one could not afford to pay more than about \$0.25/pound of fat supplement. Other possible benefits from fat supplementation such as improved health or body condition were not considered in these calculations. Certainly fat costs

would change if the same results could be achieved by feeding less fat than 0.7 to 0.8 pounds per day. If conception rate and/or milk production could be increased further than outlined here, then benefits would increase.

### **When to Initiate Fat Supplementation**

Fat feeding must be initiated long enough before the fats are needed for restoring the reproductive tissues to a new fertile state. This would involve the involution of the uterus, the return of the ovaries to growing and ovulating new follicles, and the uterus to receiving and maintaining a new embryo successfully. As will be discussed later, cows fed selected fat sources have responded with larger (still of acceptable size) ovarian follicles. Since ovarian activity usually returns within the first 4 weeks of calving, initiating fat feeding prepartum would allow the absorbed fatty acids to influence early ovarian activity. Feeding supplemental fat for at least 21 days, preferably for 40 days, prior to the desired physiological response is our recommendation. We have begun supplementing cows in the close-up nonlactating period (3 to 5 weeks before the calculated due date). This allows the tissues to begin storing the key fatty acids prior to when they will be most needed. We conducted an experiment to test whether the initiation of fat supplementation (Megalac-R) (2% of dietary dry matter) should begin at 5 weeks prepartum, at calving, or at 28 days postcalving. Cows fed fat starting in the prepartum period produced more milk and had fewer health problems in the first 10 days after calving than cows in the other groups. If some fat sources provide a benefit to the cow's immune system, then the fat feeding should begin during the transition period. In summary, research studies that have documented the benefits of fat supplements for reproduction fed the fats at a rate of at least 1.5% of the diet dry matter. Feeding fat supplements at a lower inclusion rate may prove beneficial in the field, but the research studies to support a lower feeding rate have not been done, as far as we know.

### **How Might Fat Supplementation Help Improve Conception Rates?**

Improvements in reproductive performance through dietary fat supplementation may result for a variety of feasible reasons, either acting alone or in combination. The research support behind each of these is not equal, although each has a biological basis to be true. The main hypotheses are listed below as a group and then are discussed individually in the following paragraphs.

1. The feeding of additional energy in the form of fat reduces the cow's negative energy status so that she returns to estrus earlier after calving and therefore is more fertile at insemination.
2. Feeding additional essential fatty acids in the diet cures a fatty acid deficiency that has developed in the modern-day lactating dairy cow as she is managed today.
3. Cows fed fat develop larger ovarian follicles that develop into larger corpus lutea (CL) which produce more progesterone, a hormone necessary for coordinating nutrients for the developing embryo and for maintaining pregnancy until calving.
4. Specific individual fatty acids taken up by the uterus help inhibit the production or release of prostaglandin  $F_{2\alpha}$  ( $PGF_{2\alpha}$ ) by the uterus when the embryo is ~2.5 weeks old. This helps prevent the regression of the corpus luteum on the ovary so that progesterone continues to be produced and the newly formed embryo survives.



5. The fertilization rate and embryo development is improved when fat is fed.
6. The immune status of the cow may be improved, reducing her susceptibility to disease, and improving her chances of becoming pregnant.

### **Improving Energy Status?**

Those lactating dairy cows which experience a prolonged and intense negative energy state have a delayed resumption of estrous cycles after parturition which can increase the number of days open. If fat supplementation can help increase energy intake, then possibly the negative energy state can be lessened and estrous cycles start sooner and conception occur sooner. Adding a very energy dense nutrient such as fat to the diet will usually increase the cow's energy intake.

However the energy status of the cow is usually not improved because of a slight to moderate depression in feed intake and/or an increase in milk production. Dairy cows fed tallow at 3% of dietary DM tended to have a greater pregnancy rate (62 vs. 44%; Table 2) despite having a more negative calculated mean net energy status from weeks 2 to 12 postpartum compared to cows not fed tallow. Likewise cows fed calcium salts of CLA (Castaneda-Gutierrez, 2005) or palm oil distillate (Garcia-Bojalil et al., 1998; Sklan et al., 1991) had better conception rates without an improvement in energy balance. Although there is evidence that the feeding of fat can improve the energy status of lactating dairy cows, an improvement in reproductive performance occurred in several instances apart from an improving energy status of the experimental animals.

Therefore fat supplementation likely is improving reproductive performance by other means.

### **Meeting an Essential Fatty Acid Requirement?**

Although current wisdom in the dairy industry is that the dietary intakes of linoleic and linolenic fatty acids are sufficient for meeting the lactating cow's requirements, the recently developed fat sub model of the Cornell-Penn-Miner (CPM) Institute Dairy Ration Analyzer v3.0.7a ([mail.vet.upenn.edu/~ejjancze/cpmbeta3.html](mailto:vet.upenn.edu/~ejjancze/cpmbeta3.html)) (Moate et al., 2004) indicates that the modern cow is exporting more linoleic acid in her milk than she is absorbing from her diet; that is, she is in a negative linoleic acid balance. For example, using data from a recent study at the University of Florida, the model calculated that the diet supplied 33 grams of linoleic acid but the milk put out 53 grams of linoleic acid, a 20 gram/day deficiency. The remainder must have been supplied from adipose tissue. The pools of C18:2 in adipose tissue are likely very dynamic. Feeding fat sources rich in linoleic acid that can reach the small intestine may reduce the negative balance of linoleic acid and improve performance. Nonruminant animals, such as pigs and poultry, had their reproductive performance greatly improved when an essential fatty acid deficiency was solved. Certainly the lactating cow does not show obvious signs of fatty acid deficiency such as scaly skin and dandruff so if a deficiency does exist, it is not severe.

### **Healthier Ovarian Follicles?**

In the initial days of the estrous cycle, a group of small follicles grow up on each ovary. From this group, one follicle (called the dominant follicle) continues to grow while the others disappear. This will usually happen two or three times during a single estrous cycle. These dominant follicles increase in diameter from a detectable size of 3 mm up to about 15 to 18 mm before regressing or ovulating. After the dominant follicle releases its egg into the oviduct, the

ruptured follicle forms a yellow structure called a corpus luteum, which produces the very important hormone called progesterone. Progesterone not only prepares the uterus for implantation of the embryo but helps coordinate the nutrients for development of the embryo and also maintains pregnancy by maintaining a quiet uterus until parturition. Cows that have a greater concentration of progesterone in their blood after insemination (during days 4 to 15) also have a better chance of becoming pregnant. What leads to greater progesterone in the blood? A large corpus luteum formed from a large dominant follicle that ovulated. Therefore larger dominant follicles (up to about 20 mm in size) are often beneficial. Ovulation of smaller follicles is associated with a lower conception rate.

Table 4. Diameter of the dominant ovarian follicle of lactating dairy cows fed fat supplements was greater than that of cows fed the control diet ( $P < 0.10$ ).

Reference	Fat source	Experimental diets	
		Control	Fat
		----- mm -----	
Lucy et al., 1991	Ca salt of palm oil	12.4	18.2
Lucy et al., 1993	Ca salt of palm oil	16.0	18.6
Oldick et al., 1997	Yellow grease	16.9	20.9
Beam and Butler, 1997	Tallow -Yellow grease	11.0	13.5
Staples et al., 2000	Soybean oil, fish oil	14.3	17.1
Robinson et al., 2002	Protected soybeans	13.3	16.9
Bilby et al., 2006	Megalac-R or Flaxseed oil	15.0	16.5
Ambrose et al., 2006	Rolled flaxseeds	14.1	16.9
Average		14.1	17.3

The size of the dominant follicle is often larger in lactating dairy cows receiving supplemental fat. On average, the size of the dominant follicle was 3.2 mm larger (a 23% increase) in fat-supplemented cows compared to control cows (Table 4). As shown in Table 4, a variety of dietary fat sources have had this effect on cow ovaries. Yet are certain fats more effective? Some studies did compare fat sources head-to-head. In two studies, it was the feeding of fats enriched in omega-6 (linoleic acid) or omega-3 fatty acids (linolenic or EPA and DHA) (Staples et al., 2000; Bilby et al., 2006) that stimulated larger dominant follicles compared to fats enriched in oleic acid. Thus the polyunsaturated fats were most effective in increasing follicle size. In summary, cows fed fats enriched with the essential fatty acids are likely to have more progesterone being synthesized due to larger ovarian corpus lutea, thus making them better candidates for a successful pregnancy.

## Less Embryonic Loss?

Here too, progesterone plays an important role. The embryo must signal to the uterus that it is present, so that the uterus does not release prostaglandin  $F_{2\alpha}$ . If prostaglandin  $F_{2\alpha}$  is released by the uterus, the corpus luteum will disappear, progesterone synthesis will drop, the embryo will die for lack of support, and the cow will start a new estrous cycle. About 50% of embryos die (~40% during the first 28 days after AI and ~14% between 28 and 45 days after AI). Embryonic loss is a significant problem in the dairy industry.

Omega-3 fatty acids stored in the uterus from the diet can aid the process of embryo preservation by helping to reduce the synthesis of prostaglandin  $F_{2\alpha}$ . Can omega-6 fatty acids have a similar beneficial effect? Not likely, because omega-6 fatty acids are used to synthesis prostaglandin  $F_{2\alpha}$ . As proof, lactating dairy cows fed soybeans or sunflower seeds (both good sources of linoleic acid, the omega-6 fatty acid) had increased concentrations of prostaglandin  $F_{2\alpha}$  in their blood when the uterus was artificially stimulated with an oxytocin injection. Cows that are fed omega-3 fatty acids partially replace the omega-6 fatty acids stored in the uterus so that there is less omega-6 inventory for the cow to draw from for synthesis of prostaglandin  $F_{2\alpha}$ . In demonstration of this effect, cows fed omega-3 fatty acids in the form of fish oil, flaxseed, or fish oil plus flaxseed in 4 different studies had lower concentrations of prostaglandin  $F_{2\alpha}$  in their blood when the uterus was artificially stimulated by an oxytocin injection.

If dietary omega-3 fatty acids are exerting a suppressing effect on  $PGF_{2\alpha}$  around the time of embryo recognition, then embryo loss should be reduced. Holstein cows ( $n = 121$ ) were allotted to one of two dietary treatments initiated at  $55 \pm 22$  days postpartum (Ambrose et al., 2006). Diets were isonitrogenous, isoenergetic, and isolipidic. Diets contained either rolled flaxseed (high in linolenic, omega-3) or rolled sunflower seed (high in linoleic, omega-6). Cows fed flaxseed were twice as likely to become pregnant. Embryo mortality from day 32 post AI to calving was lower for cows consuming flaxseed compared to those fed sunflower seeds (9.8 vs. 27.3%). In summary, supplementation with omega-3 fatty acids may aid in suppressing prostaglandin  $F_{2\alpha}$  to prevent regression of the corpus luteum in order to maintain progesterone synthesis and sustain pregnancy (e.g. prevent early embryonic death).

## Better Quality Embryos Produced?

All embryos are not created equal. Embryos are classified as high quality when they have a symmetrical and spherical mass with individual cells that are uniform in size, color, and density. These are most likely to become established and result in a diagnosed pregnancy. 154 California dairy cows were supplemented with either a calcium salt blend of linoleic acid and trans C18:1 (EnerG I Transition Formula<sup>®</sup>) or a calcium salt of palm oil (EnerG II<sup>®</sup>) (Virtus Nutrition) from 25 days before calving through 60 days postpartum at which time the cows underwent timed AI. Five days after AI, the uterus was flushed out to recover and evaluate the fertilized structures (Cerri et al., 2004). A greater proportion of the cows fed the mixture of linoleic acid and trans fatty acids tended to have fertilized structures compared to those fed the other fat source (87 vs. 73%), they had more sperm attached to each structure collected (34 vs. 21), and they tended to have more of their embryos classified as high quality (73 vs. 51%). In a larger set of cows numbering 397, conception rate at first AI was greater for cows fed the linoleic and trans acid

mixture (33.5 vs. 25.6%) (Juchem et al., 2004). It is not clear if linoleic acid or the trans fatty acid in this mixture was most responsible for this benefit. The fatty acids in the supplement likely changed the fatty acid makeup of the cell membranes of these structures flushed from the cow's uterus, improving their quality. In a second study, the embryos collected from superovulated Holstein cows fed whole unprocessed flaxseed and transferred to Holstein heifers resulted in a better gestation rate than embryos coming from cows fed Megalac (58.8 vs. 29.3%) (Petit et al., 2004). The diet of the donor animal was more important than the diet of the recipient animal in this study suggesting that the dietary fat helps the cow develop a robust embryo.

### **Improved Immune Status?**

During the first four weeks postpartum, the cow's immune system is severely challenged. The incidence of diseases and disorders can be high during this time and these, in turn, can have a negative impact on reproductive performance. In a study involving 2087 cows, those that had clinical mastitis during the first 45 days postpartum were at 2.7 times greater risk of abortion within the next 90 days compared to those without mastitis (Risco et al., 1999). Cows in the Netherlands were much less likely to become pregnant if they experienced a displaced abomasum or retained fetal membranes (Loeffler et al., 1999). A relatively new area of research interest is the use of nutrients for improving/maintaining animal health. As a pharmaceutical is a medicinal drug, a "nutraceutical" may be defined as a nutrient having a medicinal effect.

We are learning that the fatty acid profile of the lymphocytes (the primary cells that recognize and respond to pathogens) can be changed by the type of fat the cow consumes. This in turn can change the way animals handle an infection (Calder et al., 2002), although cows have not received much attention by scientists in this area. The lymphocytes responded better when challenged by a pathogen when cows were fed whole flaxseed (high in linolenic acid) than when fed soybeans (high in linoleic acid) (Lessard et al., 2004). In a Florida study, the lactating heifers had fewer neutrophils flushed from the uterus at ~40 days postpartum when fed flaxseed oil (1.8% of dietary DM) compared to animals fed fats enriched in oleic acid, trans fatty acids, or linoleic acid (Amaral et al., 2005). Whether these lower numbers are a sign of better health is uncertain. Neutrophils serve as the first line of defense against pathogen invaders. In a second Florida study, Holstein cows that were fed supplemental fat (2% of diet as Megalac-R enriched with linoleic acid) starting 5 weeks before their due date had fewer health problems (mastitis, retained fetal membranes, or metritis) in the first 10 days postpartum compared to cows not fed fat prepartum (1/12 vs. 15/35). These promising results need to be tested using more cows in future studies. If positive health benefits should result from fat supplementation, it seems logical to begin moderate fat supplementation in the close-up period.

### **Summary**

It has been known for many years that early postpartum dairy cows usually produce more milk when fed a moderate amount of supplemental fat. There is growing evidence, as summarized in Table 2, that lactating dairy cows can benefit reproductively as well. Fat sources enriched in omega-6 or omega-3 fatty acids that deliver these fats to tissues beyond the rumen may be the most effective ones to feed but this can not be firmly concluded because other fats having very

low amounts of these omega fatty acids have improved conception rates in single studies. The fats were fed at a minimum of 1.5% of the diet in studies in which conception rates were improved. Feeding less fat than this may be beneficial, but there is no supporting research behind it. Improved conception rates by fat-supplemented cows have been associated with an improved progesterone status of the cow by 1) increasing the size of the dominant follicle and corpus lutea on the ovaries and 2) by helping the corpus luteum survive and continue to produce progesterone during the early days of pregnancy. Early research results appear promising that some fat supplements may prove helpful to the health of the cow as well, but much more work is needed. If fed in moderate amounts, start feeding the fat when the cows enter the close-up group, especially if benefits to cow health and the ovaries are desired.

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