HIGH ENERGY INTAKE REGULATES HORMONE METABOLISM: RESULTANT CHANGES IN REPRODUCTIVE PHYSIOLOGY AND MANAGEMENT OF LACTATING DAIRY COWS

Milo Wiltbank, Hernando Lopez, Roberto Sartori, Ahmet Gument Department of Dairy Science, University of Wisconsin-Madison

wiltbank@wisc.edu

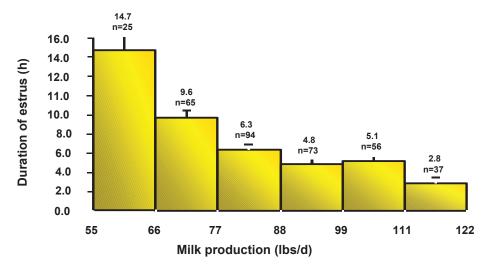
Reproductive efficiency has major impacts on profitability of livestock operations. Livestock producers have utilized many methodologies in order to improve reproductive efficiency including the use of reproductive hormones to regulate and control the estrous cycle (Yaniz et al., 2004). Nutritional strategies have also been utilized independently or in conjunction with hormonal programs to improve reproductive efficiency. However, there are numerous interactions between nutrition and reproduction that are not yet defined. For example, increasing energy consumption in dairy heifers appears to decrease embryonic survival (Dunne et al., 1999). In addition, lactating dairy cows that have very high feed consumption appear to have a reduction in reproductive efficiency. Our recent work has shown that increasing feed consumption causes reproductive hormones to be metabolized at greatly increased rates. The key hormones that are metabolized at high rates are the steroid hormones, estrogen and progesterone. It seems likely that changes in steroid metabolism could be altering reproduction in many circumstances that have not been previously recognized. This manuscript focuses on lactating dairy cows and determining which specific aspects of reproductive physiology are being altered by high steroid metabolism, defining the whole animal physiology and temporal patterns involved in acquiring this high steroid metabolism and in practically improving reproductive efficiency in these cows.

There are numerous changes in reproductive physiology that are apparent in high-producing lactating dairy cows. Time in heat is reduced in lactating cows to less than 8 h (Nebel et al., 1997). Conception rate is lower in lactating cows (generally 25-40%) than heifers (60-75%; Pursley et al., 1997). Twinning rate in dairy cows is greater than in heifers (Ryan and Boland, 1991) and can be as high as 20% in some herds. Pregnancy loss is greater in lactating cows than heifers (Santos et al., 2004). Other reproductive abnormalities have also been reported in lactating dairy cows (Lamming and Darwash, 1998; Royal et al., 2000). Changes in some of these reproductive values are associated with level of milk production. An understanding of the changes in reproduction that are occurring in high producing dairy cows will allow us to implement reproductive management programs on dairy farms that will maximize profitability.

Duration of Heat (estrus)

We have recently completed a study in which we evaluated the duration of estrus in a group of lactating dairy cows using the HeatWatch system (Lopez et al., 2004a). This system allowed continuous monitoring of all mounts on a cow 24 h per day and can be used to calculate the duration of estrus in individual dairy cows. Cows with milk production above the herd average (87 lb/d) had shorter (P < 0.001) duration of estrus (6.2 ± 0.5 h) than cows with lower milk production (10.9 ± 0.7 h). This effect of milk production was not due to a parity effect because separate analysis of primiparous and multiparous cows showed a similar effect. Figure 1 shows the relationship between level of milk production and duration of estrus.

What does this practically mean for a dairy farm? We used these data to analyze what would happen to heat detection efficiency for cows with different levels of milk production. In Fig. 2 is shown, the probability of detecting a cow in heat with different frequency of heat detection. If a cow is producing about 70 lbs of milk per day, a 4 time per day heat detection program will detect about 90% of cows that are in estrus. However, this same program (4 times/d) will only detect about 50% of cows in heat if they are producing above 100 lbs/d. This result gets even worse if heat detection is done only twice per day or once per day. It should be noted that all of the probabilities in this analysis were based on actual ovulation by the cows (detected by ultrasound). Some producers will say that the high producing cows are not cycling, but they are cycling normally; they do not detect them in heat because they have so short of a time that they are in heat. Increasing number of times that cows are checked for heat can help to solve this problem but many times synchronized ovulation programs like Ovsynch need to be implemented in order to get these high-producing cows bred in a timely manner



Analysis included all single ovulations (n=350) except first postpartum ovulations
Average milk production during the 10 days before estrus

Figure 1. Relationship between level of milk production and duration of estrus

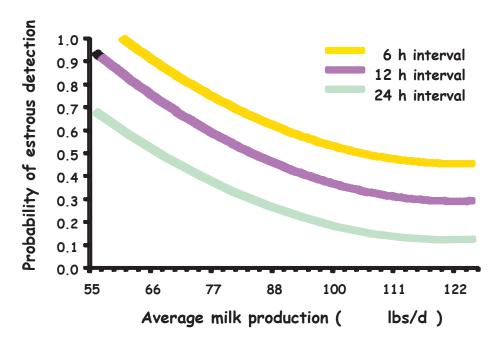


Figure 2. How the probability of heat detection changes with different frequencies of heat detection and different levels of milk production

Anovular Cows

Anestrus vs. Anovular. Cows that are not detected in heat will be, many times, assumed to not be cycling or ovulating. However, this is not the case in many instances. There are many reasons that cows may not manifest standing heat even though they have an LH surge and ovulate. Obviously, one of the critical problems is the type of footing that is provided for the cows during heat detection. In addition, the methods and amount of time spent in heat detection can result in more or fewer cows being detected in heat. As discussed above, milk production could also be causing cows to not show heat.

As shown in Fig. 3 there is essentially no relationship between level of milk production and percentage of cows that were not cycling (anovular). This means that low producing cows are just as likely to be not cycling as high producing cows. However, as mentioned above, the high producing cows may be harder to catch in heat. The other surprising finding in this study as well as many other recent studies is that about 20% of lactating dairy cows are not cycling.

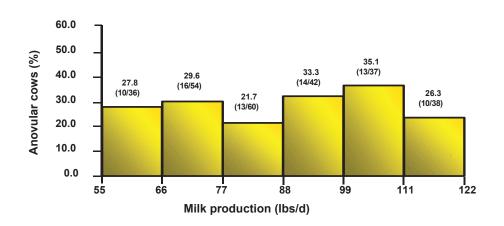


Figure 3. Lack of a relationship between milk production (50-70 d postpartum) and percentage of cows that were anovular at 70 days in milk (n = 267)

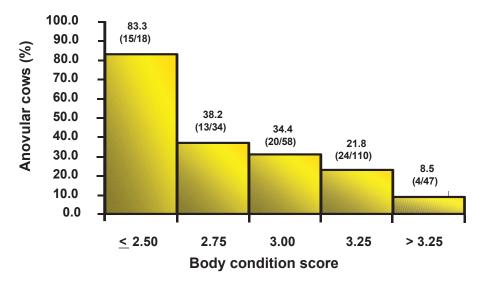


Figure 4. Relationship between body condition score (closest to 60 d postpartum) and percentage of cows that were found to be anovular at 70 days in milk (n = 267)

Body Condition Score and Anovular Cow. It has been known for some time that cows with low body condition score have a greater likelihood to not be ovulating. In Fig. 4 is shown the relationship between body condition score at day 70 after calving and percentage of cows that are anovular. It is clear that most cows that have a low body condition score at this time are anovular. Thus, of the 18 cows that had a body condition score of less than 2.5, 15 were found to be anovular. These cows generally have small follicles. However, the majority of cows that are anovular are found in the cows with good body condition score were found to be anovular. These cows generally have follicles that are larger than ovulatory size (17 mm) and usually larger than 20 mm. They also sometimes have very large follicular cysts. The intriguing thing about these data is that although clearly cows with low body condition score are likely to be anovular, most of the anovular cows in the dairy herd do not have low body condition scores.

Treatment of Anovular Cows. An obvious treatment for anovular dairy cows is to treat them with the Ovsynch protocol. Unfortunately, our results have not been as encouraging as we hoped with this protocol in anovular cows (Table 1). We performed an experiment in which we evaluated the ovaries of 316 lactating dairy cows on a commercial dairy using ultrasound (Gumen et al., 2003). After weekly evaluations we classified the cows as either ovular (cycling) or

anovular (not cycling). We then randomly assigned the cows to either receive the Ovsynch protocol or be checked for estrus during a 21 day time period. The Ovsynch protocol began on day 60 and the cows bred to heat detection (Estrous) also began to be checked for heat and inseminated after day 60. The Ovsynch cows were all bred on the 10th day of the treatment period with a timed AI. More Ovsynch cows were bred than heat detection in the ovular or anovular groups. The conception rate was similar for ovular cows that were bred to a standing heat or to the Ovsynch protocol. However, anovular cows had a very low conception rate and pregnancy rate after either reproductive management system. Thus, Ovsynch does not appear to be an effective treatment for anovular cows.

	Ovular cows			Anovular cows		
	Estrous	Ovsynch		Estrous	Ovsynch	
	n = 135	n = 117	P-value	n = 31	n = 33	P-value
Inseminated during	72 %	100 %		29 %	100 %	
21-d period	97/135	117/117	0.0001	9/31	33/33	0.0001
Double ovulation	16 %	12 %		38 %	13 %	
rate	19/120	14/114	0.4351	5/13	4/31	0.0551
Conception rate	35 %	32 %		11 %	9 %	
at ~ 60 d	34/97	37/117	0.5960	1/9	3/33	0.8547
Pregnancy rate	29 %	32 %		3 %	9 %	
at ~ 60 d	39 ^c /135	37/117	0.6370	1/31	3/33	0.3326
Embryo loss from	11 %	14 %		50 %	0 %	
28 to 64 d	4/38	6/43	0.6398	1/2	0/3	

Table 1. Comparison of ovular and anovular cows in the estrous detection vs.

 Ovsynch group

We (unpublished results) have evaluated if combining a CIDR with Ovsynch would be a more effective treatment for anovular cows. In this experiment 634 cows were evaluated to determine which cows were anovular. This was done by taking blood samples 10 days apart. If progesterone was low in both blood samples then the cows were designated as anovular. All cows were randomly assigned to either receive Ovsynch or receive Ovsynch with a CIDR inserted from the first GnRH until the time of $PGF_{2\alpha}$. This protocol has sometimes been designated as CIDR-Synch. Overall, the cows that were treated with the CIDR-Synch program had a 42% conception rate as compared to a 32% conception rate for the Ovsynch cows. However, if we evaluated only the cows that were ovular before Ovsynch, then Ovsynch and CIDR-Synch gave similar results. The CIDR-Synch program primarily improved reproduction in the noncycling cows. There were 24.4% of the cows that were non-cycling (anovular) in this study. If anovular cows were treated with Ovsynch alone then 22.2% got pregnant. However, if anovular cows were treated with CIDR-Synch then 36% of cows became pregnant. This was similar to the conception rate in ovular cows (39%). Thus, the CIDR-Synch program seems to work better than the Ovsynch program alone in anovular cows but does not appear to improve conception rates in ovular cows.

Double Ovulation Rate

Another reproductive trait that has been directly linked to milk production is double ovulation rate. From a practical standpoint double ovulation rate appears to be the underlying cause of increased twinning rate in lactating dairy cows with 93% of twins being non-identical (Silva Del Rio et al., 2004). Numerous factors have been recognized as possible regulators of twinning rates including: age of dam, season, genetics, use of reproductive hormones or antibiotics, ovarian cysts, days open, and peak milk production (reviewed in Wiltbank et al., 2000). In a large study on risk factors for twinning, Kinsel et al. (1998) concluded, "the single largest contributor (>50%) to the recent increase in the rate of twinning is the increase in peak milk production". We performed a study in which we evaluated double ovulation rate in 240 dairy cows (Fricke and Wiltbank, 1999). All cows had ovulation synchronized with the Ovsynch protocol (Pursley et al., 1995) that uses two treatments with GnRH and one treatment with $PGF_{2\alpha}$. Ovulation was evaluated by transrectal ultrasound at the time of the second GnRH injection and 48 h after this injection. The mean milk production was determined 3 d before ovulation and averaged 90 ± 1.8 lbs/d. The cows were segregated by whether they were below or above the mean value. Double ovulation rate in cows that were high milk producers was 20.2% compared to 6.9% in low producers (P < 0.05). This difference was similar regardless of lactation number. We (Lopez et al., 2005) have also found a similar relationship between milk production and double ovulation rate in naturally ovulating cows (Fig. 5). Cows that produced less than 88 lbs/d had a very low double ovulation rate; whereas, cows above 111 lbs/d had more than a 50% double ovulation rate. This is an incredible difference in double ovulation rate and will clearly impact the twinning rate in these cows. It should be remembered that this effect of milk production is not due to the total milk production during the entire lactation but is most related to the milk production within the 2 weeks before the animal came into heat.

From a practical standpoint, it appears that there may be little that we can do to change this trend. Using Ovsynch does not seem to increase or decrease double ovulation with double ovulation related to milk production whether we look after a hormonal synchronization program or a natural estrus. Obviously, not all double ovulations result in twins but increasing double ovulation rate will almost surely result in increased twinning rates on higher producing farms. It seems clear that the main increase occurs after cows are producing about 90 lbs/d. Thus, we must anticipate that we will have a dramatic increase in double ovulation rate in cows producing over 90 lbs/d and this will result in an increase in twinning rate in cows that conceive during this time of high milk production. We must align our management procedures to deal with this increasing twinning rate if we are increasing milk production into this range in our dairy. First, we must set a program to **diagnose** twins. Second, we should set up procedures to **manage** cows that are likely to have twin births. Twinning cows will calve earlier (10-14 d on average) and are likely to have more problems during the calving process. These twin calving cows were, on average, our highest producing cows during the previous lactation, therefore, we must carefully design our calving and early lactation procedures with these twinning cows in mind.

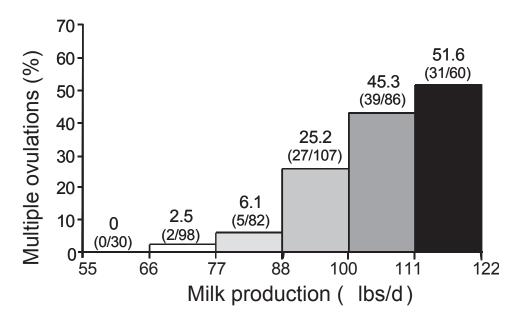


Figure 5. Incidence of multiple ovulation vs. milk production during the 14 d before estrus

Fertility and Early Embryonic Development

The relationship between various measures of fertility (conception rate) and level of milk production remains controversial. Washburn et al. (2002) analyzed the relationship of conception rate and milk production over more than a 20 year time period (1976-1999) in dairy herds in the Southeastern U.S. It was clear that conception rates decreased from about 55% to about 35% during this time period as milk production dramatically increased. Faust et al. (1988) showed a clear relationship between level of milk production and conception rate in primiparous Holstein dairy cattle. In contrast, Peters and Pursley (2002) reported that higher producing cows had greater conception rates following Ovsynch than lower producing cows. Obviously, fertility is a complex trait and is likely to be related to numerous factors including: uterine infection, negative energy balance, urea concentrations in the blood, vitamins, fertility of sire, accuracy of heat detection, insemination technique, etc. (Gröhn and Rajala-Schultz; 2000; Lucy, 2001). For example, an increase in double ovulation rate in high-producing dairy cows (illustrated above) would increase the chances for pregnancy even though possible negative effects of high milk production could decrease the percentage of ovulated oocytes that produce a pregnancy. Thus, a simple relationship between milk production and conception rates seems unlikely.

We have been interested for a number of years in the underlying mechanisms that produce the lower fertility in lactating dairy cows. In two recent experiments we tested the hypothesis that lactating dairy cows have reduced fertilization rate and early embryonic development compared to non-lactating females during normal reproductive cycles (NOT superovulated; Table 2; Sartori et al., 2002b). Experiment 1 compared lactating Holstein cows (n = 27; 70-140 d postpartum; 40.0 ± 1.5 kg milk/d) to nulliparous heifers (n = 28; 11-17 mo old) during summer, and Exp. 2 compared lactating cows (n = 27; 37-60 d postpartum; 45.9 ± 1.4 kg milk/d) to dry cows (n = 26) during winter. Cows had AI at estrus with combined semen from four high fertility bulls. Embryos or oocytes were recovered 5 d after ovulation and evaluated for fertilization, embryo quality (1 = excellent to 5 = degenerate), number of nuclei/embryo, and number of accessory sperm. An improved embryo flushing technique increased recovery rate of embryos/oocytes in Exp. 2 than in Exp. 1. Fertilization rate was lower in lactating cows in the summer experiment but was not reduced during the winter. This reduction appeared to be due to a heat stress effect on the oocyte because 80% of the unfertilized oocytes (UFOs) had sperm attached with an average of $17.8 \pm$ 12.1 sperm/UFO. Although heifers were exposed to a similar heat stress, their increase in body temperature was minimal as compared to a large increase in body temperature in lactating cows. Embryo quality was reduced in lactating

dairy cows during either summer or winter. For example, the percentage of viable embryos (Grade 1-3) was reduced from 82% in non-lactating cows to 53% in lactating cows. Thus, embryos of lactating dairy cows were detectably inferior to embryos from non-lactating cows as early as 5 d after ovulation with about 50% non-viable embryos.

Table 2. Summary of embryo results comparing heifers versus lactating cows during summer (Experiment 1) or non-lactating (Dry) cows versus lactating cows during winter (Experiment 2)

	Experiment	1 (summer)	Experiment 2 (winter)	
	Heifers	Lactating cows	Dry cows	Lactating cows
Recovery rate per CL, %	39.5	30.9	55.9	60.3
(no. embryos or UFOs/no. CL)	(32/81)	(38/123)	(38/68)	(41/68)
Fertilization rate, %	100b	55.3a	89.5	87.8
(no. embryos/no. structures*)	(32/32)	(21/38)	(34/38)	(36/41)
Embryo quality, mean ± SEM	$2.2 \pm 0.3a$	$3.8 \pm \mathbf{0.4b}$	2.2 ± 0.3 c	$3.1 \pm 0.3 d$
Nuclei/embryo, mean \pm SEM	$36.8 \pm 3.0b$	$19.3 \pm 3.7a$	30.6 ± 2.1	27.2 ± 2.7
Grade 1-3 embryos, %	71.9b	33.3a	82.3b	52.8a
(no./no. embryos**)	(23/32)	(7/21)	(28/34)	(19/36)

a,bDifferent within row within exp.; P < 0.05.

c,dDifferent within row within exp.; P = 0.06.

*Total number of structures (embryos/UFOs) recovered.

**Total number of embryos recovered.

From a practical viewpoint, it appears that many of the problems with fertility in dairy cows appear to occur during the first week after breeding. We hypothesized that we could improve reproduction just by transferring a good quality embryo at 7 days after expected time of AI. So in a fairly large experiment we compared conception rate (CR) in our herd when cows were bred either by AI or by embryo transfer (ET). During 365 d, 550 potential breedings were used from 243 lactating Holstein cows (77 lbs of milk/d). Cows were synchronized (GnRH-7d-PGF_{2 α}-3d-GnRH) and randomly assigned to receive AI immediately after the second GnRH injection (d 0) or to receive transfer of one embryo 7 d later. Circulating progesterone and follicular and luteal size were determined on d 0 and 7. Pregnancy diagnosis was performed on d 25 or 32, and pregnant cows were reevaluated on d 60-66. Synchronized cows with single ovulation had similar (P > 0.30) CR on d 25-32 with ET (n=176; 40.3%) and AI (n=160; 35.6%). Pregnancy loss between d 25-32 and 60-66 also did not differ (P = 0.38) between ET (26.2%) and AI (18.6%). When single (n=334) and multiple (n=57) ovulators were compared, independent of treatment, multiple ovulators

had greater (P < 0.001) circulating progesterone on d 7 (2.7 vs. 1.9 ng/ml) and there was a tendency (P = 0.10) for greater CR in multiple ovulators (50.9% vs. 38.1%). However, there was no difference in CR between AI and ET cows with multiple ovulation (50.0% vs. 51.7%). The CR tended to be lower for AI than ET in single-ovulatory cows ovulating smaller (≤ 15 mm; 23.7 vs. 42.3%; P = 0.06) but not average (16-19 mm; 41.2 vs. 37.3%; P = 0.81) or larger (≥ 20 mm; 34.3 vs. 51.0%; P = 0.36) follicles. Thus, ET did not improve overall CR in lactating cows but size and number of ovulating follicles may determine success with these procedures. We obviously have a large number of future experiments to do in order to resolve the problems with fertility in lactating dairy cows.

Steroid Metabolism in Lactating Dairy Cows

The mechanisms that produce these many changes in reproductive physiology in lactating cows have not yet been defined. The next two sections will provide our current ideas about one reason that reproductive changes that are closely associated with milk production (such as duration of heat, double ovulation rate) are occurring and where we are going with research to improve these problems. Concentrations of circulating steroid hormone (estrogen - E₂ and progesterone - P_4) are involved in almost every aspect of reproductive physiology. Circulating concentrations of hormones, including steroids, are determined by rate of production and rate of metabolism of the hormone. Changes in metabolism of steroid hormones due to an increase in feed consumption, such as during lactation, can dramatically alter circulating P₄ during continuous delivery of P₄ (Parr et al., 1993; Rabiee et al., 2001a; 2001b). Alterations in steroid metabolism could alter the reproductive physiology of any species, but may particularly alter reproduction in species with extreme increases in feed intake such as lactating dairy cows. We propose that some reproductive changes in lactating dairy cows are caused by dramatic increases in steroid metabolism due to elevations in feed consumption and liver blood flow.

In a recent series of experiments we tested the hypothesis that increased liver blood flow (LBF) as a result of elevated feed intake in lactating dairy cows would increase steroid metabolism (Sangsritavong et al., 2002). We found that prior to feeding, the LBF was greater in lactating (1561 ± 57 l/h) than similar size and age non-lactating (747 ± 47 l/h) cows. The LBF and metabolism of progesterone and estrogen increased immediately after any amount of feed consumption in both lactating and non-lactating cows (Sangsritavong et al., 2002). The metabolism of estrogen and progesterone was much greater ($2.3 \times$) in lactating than non-lactating cows. Thus, the changes in metabolism of estrogen and progesterone in response to feeding are immediate and appear to be related to acute changes in LBF. In lactating cows, a continuous high plane of nutrition appears to chronically elevate LBF and metabolism of these hormones to about double the amount observed in similar size and age non-lactating cows. These results indicate that even with a similar level of hormone production, there would be lower circulating hormone concentrations in lactating dairy cows. This may be the underlying physiological basis for reduced expression of estrus, increased double ovulation rate, and possibly other reproductive changes in lactating cows.

Working Model

We have synthesized this information into a working model (Fig. 6). Lactating cows have greater energy requirements than non-lactating cows (for example, a cow producing 110 lbs/d of milk will require 53 Mcal/d of energy vs. 12.5 Mcal/d for a non-lactating cow; NRC, 2001). The high feed consumption required to meet these energy requirements leads to a dramatic increase in liver blood flow (Sangsritavong et al., 2002). An elevation in liver blood flow leads to elevated metabolism of both estrogen and progesterone. This would cause a reduction in circulating estrogen and progesterone concentrations even in the midst of high production of steroid hormones by the follicle or corpus luteum.

This simple model could potentially explain some of the results described in the sections above. For example, high estrogen concentrations cause a cow to come into heat. In lactating dairy cows estrogen reaches lower concentrations and decreases faster after the cow shows heat. Therefore, it makes sense that a higher producing cow would have a shorter heat because estrogen is being metabolized at a higher rate in high producing than lower producing cows. Obviously, the critical involvement of estrogen and progesterone in almost every aspect of reproductive physiology makes changes in steroid metabolism an extremely attractive explanation for the numerous changes in reproduction that have been observed in lactating dairy cows. Nevertheless, it is possible that only a few or none of these reproductive changes are related to increased steroid metabolism in lactating dairy cows. Our research continues to focus on expanding and testing this working model. There are two key steps shown below that we are trying to change in our experiments. We are testing different feeds or treatments that could block metabolism of estrogen and progesterone (steroid metabolism). This could eliminate the problems associated with steroid metabolism in lactating dairy cows and possibly lead to improved reproduction. We have no inhibitors that are currently ready for use on dairy farms. Secondly, we are testing whether we can supplement estrogen and/or progesterone to improve reproduction. This could

overcome these problems by directly adding the needed steroid hormones. At this point we know that we can improve expression of heat but we have not yet found a protocol that can be used to improve fertility. These experiments are ongoing.

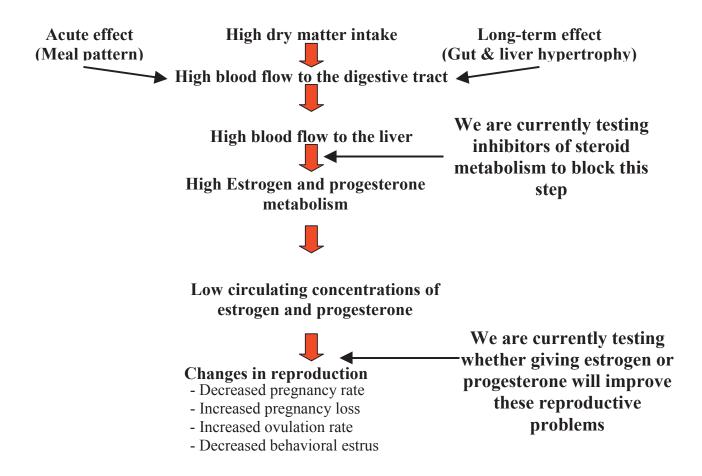


Figure 6. Schematic of the potential physiological pathway that may produce the changes in reproductive physiology observed in high-producing lactating dairy cows

Implications

This article has provided some information on how reproduction is changing in lactating dairy cows. We now have methods to directly implement on dairy farms to overcome some of these problems. The problem with duration of estrus is likely a problem that is directly related to metabolism of estrogen in high producing dairy cows. Implementation of timed AI synchronization protocols such as Ovsynch can decrease the dependence of dairy producers on detection of estrus. These are currently being very effectively utilized on many dairies with high producing dairy cows. Supplementing synchronization protocols with estrogen treatments can dramatically increase expression of estrus during this protocols (35% vs. 85%; Wiltbank, unpublished). Nevertheless, expression of estrus alone does not increase the fertility associated with timed AI programs. An important implication of this problem is that the highest producing dairy cows in a herd will be the least likely to be detected in estrus and bred in a timely manner. This is not due to lack of cyclicity in these cows (anovulation) but is due to a dramatically reduced duration of estrus expression.

Most dairies will have about a 20% incidence of anovular cows at the time breeding programs are implemented (~70 d postpartum). Some of the these cows will have low body condition scores (~one third) but most will have normal body condition and large follicles on the ovaries (~ two-thirds of anovular cows). These cows can be treated with Ovsynch but newer methods incorporating CIDR into the Ovsynch appear to produce higher conception rates in lactating cows. Cows with low body condition scores need to have improved energy balance before fertile cyclicity will return. However, there are currently no scientifically valid methods to effectively prevent the anovulation that occurs in cows with good body condition scores, representing the majority of anovular cows. At the present time, hormonal treatment methods are the most effective way to manage these type of cows. Currently used methods that are effective include: Ovsynch, Ovsynch with CIDR, or Ovsynch supplemented with estrogen (not currently approved in USA).

For twinning cows, it is clear that we know what is the primary cause of our increasing twinning rate (increasing milk production) and this is probably due to the increasing feed consumption of high producing dairy cows. Unfortunately, preventative measures are difficult because high feed consumption and high milk production are so intricately linked and in addition high feed consumption (high digestive tract and liver blood flow) and high steroid metabolism also seem to be physiologically connected. We do not currently have methods to decouple high steroid metabolism from high milk production. Therefore, cows that are producing more than 90 lbs of milk per day begin to have dramatic increases in their double ovulation rate and this translates into a dramatically increased twinning rate. As our producers are breeding cows with higher milk production (>90 lbs/d) we need to help these producers design management programs to deal with twinning cows. First, twinning cows must be identified. Second, highenergy diets must be implemented earlier during the dry period (twinner cows calve about 10 d earlier than cows calving singletons). Perhaps these twinbearing cows should not receive a traditional dry period. Notwithstanding, we now need to align our management programs to efficiently work with these twinner cows in herds with cows producing above 90 lbs/d at the time of breeding.

The problem of fertility in lactating dairy cows is extremely complex and is unlikely to be solved with a single solution on all dairy farms. Metabolism of estrogen and progesterone may be part of the underlying physiology that is producing low fertility in lactating dairy cows but clearly many other problems can be limiting in individual herds.

Literature Cited

- Dunne, L. D., M. G. Diskin, M. P. Boland, K. J. O'Farrell, and J. M. Sreeman. 1999. The effect of pre- and post-insemination plane of nutrition on embryo survival in beef heifers. Anim. Sci. 69:411-417.
- Faust, M. A., B. T. McDaniel, O. W. Robison, and J. H. Britt. 1988. Environmental and yield effects on reproduction in primiparous Holsteins. J. Dairy Sci. 71:3092-3099.
- Fricke, P. M., and M. C. Wiltbank. 1999. Effect of milk production on the incidence of double ovulation in dairy cows. Theriogenology 52:1133-1143.
- Gröhn, Y. T., and P. J. Rajala-Schultz. 2000. Epidemiology of reproductive performance in dairy cows. Anim. Repro. Sci. 60-61:605-614.
- Gumen, A., J. N. Guenther, and M. C. Wiltbank. 2003. Follicular size and response to Ovsynch versus detection of estrus in lactating dairy cows. J. Dairy Sci. 86:3184-3194.
- Kinsel, M. L., W. E. Marsh, P. L. Ruegg, and W. G. Etherington. 1998. Risk factors for twinning in dairy cows. J. Dairy Sci. 81:989-993.
- Lamming, G. E., and A. O. Darwash. 1998. The use of milk progesterone profiles to characterise components of subfertility in milked dairy cows. Anim. Repro. Sci. 52:175-190.
- Lopez, H., L.D. Satter, M.C. Wiltbank, 2004. Relationship between level of milk production and estrous behavior of lactating dairy cows. Ani. Reprod. Sci. 81:209-223.

- Lopez, H., D.Z. Caraviello, L.D. Satter, P.M. Fricke, and M.C. Wiltbank. 2005. Relationship between level of milk production and multiple ovulations in lactating dairy cows. J. Dairy Sci. 88:2783-2793.
- Lucy, M. C. 2001. Reproductive loss in high-producing dairy cattle: where will it end? J. Dairy Sci. 84:1277-1293.
- Nebel, R.L., S.M. Jobst, M.B.G. Dransfield, S.M. Pandolfi, and S.M. Bailey. 1997. Use of a radio frequency data communication system, HeatWatch, to describe behavioral estrus in dairy cattle. J. Dairy Sci. 80 (Suppl. 1):179.
- Parr, R.A., I.F. Davis, M.A. Miles, and T.J. Squires. 1993. Liver blood flow and metabolic clearance rate of progesterone in sheep. Res. Vet. Sci. 55:311-316.
- Peters, M. W., and J. R. Pursley. 2002. Fertility of lactating dairy cows treated with Ovsynch after presynchronization injections of $PGF_{2\alpha}$ and GnRH. J. Dairy Sci. 85:2403-2406.
- Pursley, J. R., M. R. Kosorok, and M. C. Wiltbank. 1997. Reproductive management of lactating dairy cows using synchronization of ovulation. J. Dairy Sci. 80:301-306.
- Pursley, J. R., M. O. Mee, and M. C. Wiltbank. 1995. Synchronization of ovulation in dairy cows using $PGF_{2\alpha}$ and GnRH. Theriogenology 44:915-923.
- Rabiee, A.R., K. L. Macmillan, and F. Schwarzenberger. 2001. Excretion rate of progesterone in milk and faeces in lactating dairy cows with two levels of milk yield. Reprod. Nutr. Dev. 41:309-319.
- Royal, M. D., A. O. Darwash, A. P. F. Flint, R. Webb, J. A. Woolliams, and G. E. Lamming. 2000. Declining fertility in dairy cattle: changes in traditional and endocrine parameters of fertility. Anim. Sci. 70:487-501.
- Ryan, D. P., and M. P. Boland. 1991. Frequency of twin births among Holstein-Friesian cows in a warm dry climate. Theriogenology 36:1-10.
- Sangsritavong, S., D. K. Combs, R. Sartori, and M. C. Wiltbank. 2002. High feed intake increases blood flow and metabolism of progesterone and estradiol-17ß in dairy cattle. J. Dairy Sci. 85:2831-2842.
- Santos, J.E.P., W.W. Thatcher, R.C. Chebel, R.L.A. Cerri, K.N. Galvao. 2004. The effect of embryonic death rates in cattle on the efficacy of estrus synchronization programs. Animal Reproduction Science 82-83(Special Issue SI):513-535.
- Sartori, R., R. Sartor-Bergfelt, S. A. Mertens, J. N. Guenther, J. J. Parrish, and M. C. Wiltbank. 2002b. Fertilization and early embryonic development in heifers and lactating cows in summer and lactating and dry cows in winter. J. Dairy Sci. 85:2803-2812.
- Silva Del Rio, N., B. W. Kirkpatrick, and P. M. Fricke. 2004. Observed frequency of monozygotic twinning in lactating Holstein cows. J. Dairy Sci. 87(Suppl. 1):65(Abstr.).

- Washburn, S. P., W. J. Silvia, C. H. Brown, B. T. McDaniel, and A. J. McAllister. 2002. Trends in reproductive performance in southeastern Holstein and Jersey DHI herds. J. Dairy Sci. 85:244-251.
- Wiltbank, M. C., P. M. Fricke, S. Sangritasvong, R. Sartori, and O. J. Ginther. 2000. Mechanisms that prevent and produce double ovulations in dairy cattle J. Dairy Sci. 83:2998-3007.
- Yaniz, J.L., K. Murugavel, F. Lopez-Gatius. 2004. Recent developments in oestrous synchronization of postpartum dairy cows with or without ovarian disorders. Reprod. Dom. Animals 39:86-93.