Five Keys for Reproductive Success

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Introduction

The efficient production of milk drives the profitability of the dairy enterprise. Within a dairy herd, total milk production is determined by the proportion of cows producing milk at any given time and the level of milk production of the individual cows within the herd (Vandehaar, 1998). Both of these factors are dramatically affected by reproductive efficiency. Improving reproductive efficiency improves profitability by maintaining the greatest proportion of cows in a herd producing milk at optimal levels.

The rate at which cows become pregnant in a dairy herd is measured using the **21-day pregnancy risk** which is determined by an interaction between service risk and conception risk. Although 21-day pregnancy risk is not necessarily the mathematical product of conception rate and service rate, this equation approximates the 21-day pregnancy risk in large groups of cows. Thus, maximizing both conception and service risk provides opportunities for management control of reproduction and profitability in a dairy operation. A practical method for determining pregnancy risk is to observe the number of successful outcomes (pregnancies) that occur during periods when eligible cows are at "risk" to become pregnant (21-day reproductive cycles). Large dairies use computer software programs to monitor reproductive performance by calculating pregnancy, service, and conception risks on a daily, weekly, or monthly basis. Pregnancy risk for lactating dairy cows in the upper Midwest region of the United States can vary between 5% to 30% among farms depending on the service and conception rate and average about 14% (Rapnicki et al., 2001). Most dairies set a reproductive goal of achieving a 20% or greater annualized 21-day pregnancy risk for their herd.

Reproductive inefficiency in lactating dairy cows not only is a source of frustration to dairy producers and their consultants but also substantially reduces dairy farm profitability. Artificial insemination (**AI**) is one of the most important agricultural technologies developed during the past century, and most dairy farms use AI to remain competitive in the current economic climate of the dairy industry. Reproductive inefficiency in lactating dairy cows, however, substantially reduces the impact and efficiency of AI. Dairy cow fertility to AI commonly is measured by calculating the percentage of cows that conceive after a single AI service, also known as the conception rate or **conception risk**. Four general factors that determine conception risk in a dairy herd include: 1) cow fertility; 2) bull fertility; 3) accuracy of heats; and 4) AI efficiency. Cow fertility refers to any cow-related factors that influence establishment of pregnancy and include factors such as inadequate nutrition and environmental stresses. Bull fertility refers to the quality of semen used for AI. Accuracy of heats refers to the timing of AI relative to estrus or ovulation, and AI efficiency refers to factors affecting pregnancy rates due to AI technique. Of these four factors, accuracy of heats and AI efficiency can be maximized through careful reproductive management practices and are further discussed below. Management strategies should be developed to maximize conception risk while realizing that high-producing herds under excellent management will likely not exceed an overall conception risk of 50%.

The 5 keys to reproductive success focus on management areas that can help to improve the overall 21-day pregnancy risk in a dairy herd.

The 5 keys to reproductive success are:

- **Key 1**: Inseminate cows quickly after the end of the voluntary waiting period
- **Key 2**: Inseminate cows at the correct time in relation to estrus or ovulation
- **Key 3**: Improve AI Efficiency
- **Key 4**: Identify nonpregnant cows early after an insemination (but not too early)
- **Key 5**: Aggressively re-inseminate nonpregnant cows

Key 1: Inseminate cows quickly after the end of the voluntary waiting period.

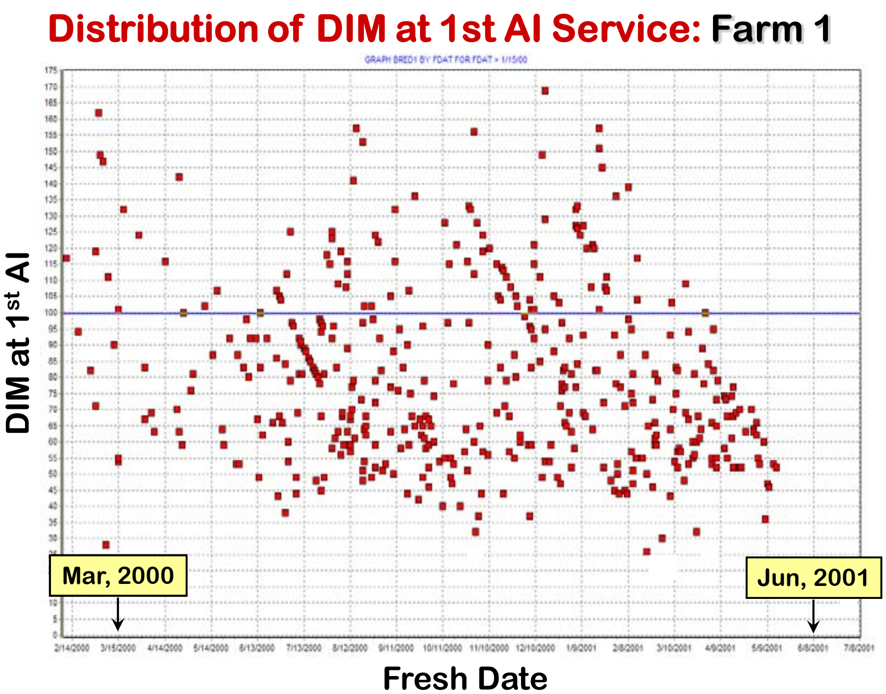
It is a fundamental principle of reproductive biology that inseminating a cow is the first step toward establishing a pregnancy. Unfortunately, on many farms a significant proportion of cows do not receive their first postpartum AI service until after 100 days in milk. First postpartum AI service represents a unique opportunity for reproductive management of lactating dairy cows because all cows in the herd have a known pregnancy status at this time (e.g., nonpregnant), which allows for use of hormonal synchronization systems that use $PGF_{2\alpha}$ without the risk of aborting a previously established pregnancy. Furthermore, reducing the interval from calving until first AI service for all cows in the herd has a profound effect on reproductive efficiency. The interval that must elapse from calving until a cow is eligible to receive her first AI service is termed the Voluntary Waiting Period (**VWP**). As the name implies, the duration of this interval is voluntary (i.e., a management decision) and traditionally varies from 40 to 70 days on most dairies.

To illustrate the advantages of programming cows to receive first AI service, we will compare reproductive data from two dairy farms in Wisconsin that employ two different strategies to initiate first postpartum AI service (Figure 1). For both graphs, days in milk (DIM) at first breeding is plotted on the vertical axis (y-axis) and time is plotted on the horizontal axis (x-axis). Each square represents an observation, or a cow within the herd, and a line has been drawn horizontally at 100 DIM. In both plots, cows receiving first AI service before 100 DIM fall below the line, whereas cows receiving first AI service after 100 DIM fall above the line. The upper plot in Figure 1 shows the pattern of cows receiving first AI service for cows in a herd managed using visual detection of estrus for first postpartum AI service, whereas the lower plot shows the pattern of cows receiving first AI service for cows managed in a herd that uses the detection of estrus combined with a Presynch-Ovsynch protocol (a hormonal synchronization system for TAI) system for first postpartum AI service.

Nearly one-third of the cows in the herd shown in the upper panel of Figure 1 exceed 100 DIM before first AI service. It should be obvious that none of these cows has a chance of becoming pregnant before 100 DIM because they have not yet been inseminated. Although most dairy producers identify a set duration for the VWP, breeding decisions for individual cows often occur before the VWP elapses. The VWP for the farm illustrated in the upper panel of Figure 1 is

50 DIM; however, many cows are submitted for AI before this time. The decision to AI a cow for the first time postpartum is determined based on when (or if) a cow is detected in estrus rather than on a predetermined management decision. In such instances, the cow is managing the decision to inseminate rather than the manager. The decision to inseminate a cow before the VWP elapses is motivated by one factor, and that factor is fear. Most producers fear the decision to not breed a cow detected in estrus because she may not be detected in estrus again until much later in lactation. Unfortunately, this risk is often realized on dairies that rely on visual estrus detection for AI because of poor estrus detection by dairy personnel and poor estrus expression by lactating dairy cows.

If the upper graph reflects the reproductive performance to first AI on your farm, you should consider using a synchronized breeding program to initiate first postpartum AI service. Use of a program such as Presynch/Ovsynch for initiating first AI service exposes all cows in the herd to the risk of becoming pregnant at or very near the end of the VWP (Navanukraw et al., 2004). In the lower panel of Figure 1, nearly all cows receive their first postpartum AI service between 65 and 73 DIM. In this scenario, the end of the VWP is roughly equal to the average day at first service for the entire herd.



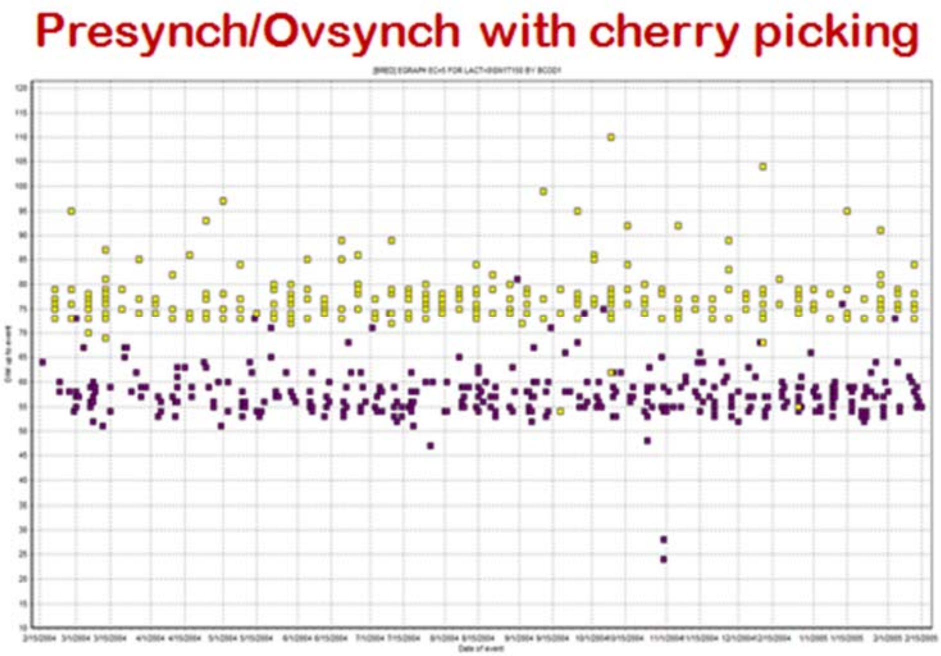


Figure 1. Days in milk at first breeding (y-axis) by time (x-axis) for cows managed using visual detection of estrus for first postpartum AI service (upper panel) and cows managed using Presynch-Ovsynch and timed artificial insemination for first postpartum AI service (lower panel).

Submission of cows for first postpartum AI using Presynch-Ovsynch

The first results with Ovsynch (Pursley et al., 1995) indicated that all nonpregnant cows could be enrolled into the protocol regardless of their stage during the estrous cycle. Subsequent results from Vasconcelos et al. (1999) using lactating dairy cows, and those of Moreira et al. (2000a) using dairy heifers showed that initiation of Ovsynch between days 5 to 12 of the estrous cycle may result in improved conception rate over the original Ovsynch protocol. Presynchronization of cows to group randomly cycling cows to initiate Ovsynch between days 5 to 12 of the estrous cycle can be accomplished using two injections of $PGF_{2\alpha}$ administered 14 days apart before initiation of the first GnRH injection of Ovsynch. A presynchronization strategy in which two injections of $PGF_{2\alpha}$ administered 14 days apart preceded initiation of Ovsynch by 12 days improved conception rates in lactating dairy cows compared to Ovsynch alone (Moreira et al., 2000b). This presynchronization strategy has become known as **Presynch-Ovsynch** (Table 1). Lactating dairy cows were randomly assigned to receive Ovsynch (n=262) or Presynch (n=264) for their first postpartum TAI, which was conducted 16 h after the second GnRH injection. The first and second $PGF_{2\alpha}$ injections for Presynch cows were administered at 37 and 51 days in milk, respectively, and all cows received a TAI at 73 days in milk. One possible hormone injection and timed AI schedule based on this research is shown in Table 1. For cycling cows, conception rate increased from 29% for Ovsynch to 43% for Presynch cows; however, no statistical treatment difference was detected when all cows (cycling and anovular) were included in the analysis). Thus, use of Presynch for programming lactating dairy cows to receive their first postpartum TAI can improve first service conception risk in a dairy herd.

Table 1. Possible hormone injection and timed artificial insemination schedule for the Presynch-Ovsynch protocol based on the results of Moreira et al., 2000b.

Sunday	Monday	Tuesday	Wednesday	Thursday	Friday	Saturday
			PGF			
			PGF			
	GnRH					
	PGF		**GnRH**	**TAI**		

PGF = prostaglandin $F_{2\alpha}$; **GnRH** = gonadotropin-releasing hormone; **TAI** = timed artificial insemination.

Key 2: Inseminate cows at the correct time in relation to estrus or ovulation.

Genetic improvement of dairy cattle through widespread use of AI has been one of the greatest success stories in all of agriculture. Before the genetic impact of AI could be realized, however, widespread adoption of this technology first had to occur. Because AI was a new technology, a simple and effective recommendation for timing of AI in relation to a detected estrus had to be developed. One of the earliest experiments (if not the earliest) conducted to establish the optimal timing of AI in relation to behavioral estrus was conducted in the 1940's and indicated that the optimal time of AI occurred about 6 to 12 h after the onset of behavioral estrus (Table 2). Unfortunately, these data were not analyzed statistically and suffer from too few cows per treatment for valid statistical comparisons. Nonetheless, a recommendation was developed in which dairy producers were instructed to inseminate cows first detected in estrus in the morning (i.e. the a.m.) later that day (i.e., the p.m.), whereas cows first detected in estrus in the evening (i.e., the p.m.) should be inseminated the next morning (i.e., the a.m.). This method has been termed the a.m./p.m. rule, and many dairy producers continue to inseminate their cows according to this rule to this day.

Table 2. Effect of time of AI on fertility of beef cows (adapted from Trimberger and Davis, 1943)

Time of AI	Number of breedings	Conception rate (%)
Start of estrus	25	44
Middle of estrus	40	82
End of estrus	40	75
Hours after the end of estrus		
6	40	36
12	25	32
18	25	28
24	25	12
36	25	8
48	25	0

From a practical perspective, the a.m./p.m. rule provided dairy producers with an easy to remember and straightforward recommendation for timing of AI in relation to behavioral estrus. Logistically, however, the a.m./p.m. rule has caused problems for the AI industry as the dairy industry has grown. Farms relying on professional AI technicians to inseminate their cows may require twice-daily visits to farms to comply to the a.m./p.m. rule, often times to breed only a single cow. Studies comparing management strategies for timing of AI conclude that once-daily

AI programs result in conception rates similar to that of using the a.m./p.m. rule (Gonzalez et al., 1985; Nebel et al., 1994; Graves et al., 1997). The data in Table 3 clearly show in a large number of cows that the 75 d nonreturn rate (a proxy for conception risk) did not differ when comparing use of the a.m./p.m. rule to a once-daily AI program. Thus, strict adherence to the a.m./p.m. rule is not required for managing a successful and effective AI program.

Table 3. Effect of once-daily AI versus the a.m./p.m. rule on 75 d nonreturn rate in lactating dairy cows (adapted from Nebel et al., 1994).

AI method	Number of breedings	75 d nonreturn rate (%)
a.m./p.m. rule	3659	60.1
Once-daily	3581	60.6

Timing of AI relative to detection of estrous behavior can also affect subsequent fertility. Based on 7,240 first service Holstein cows inseminated by professional AI technicians (Nebel et al., 1994), cows inseminated within 0 to12 h of a detected estrus had greater 75 d nonreturn rates compared to cows inseminated 12 h or more after a detected estrus (Table 4). Thus, strict adherence to the a.m./p.m. rule combined with infrequent detection of estrus on a dairy may actually result in cows receiving insemination too late relative to the timing of AI resulting in the greatest fertility (Table 4). Results reported by Nebel et al. (1994) agree with a previous recommendation that a single mid-morning AI for all cows detected in estrus the night before or the same morning should result in near optimal conception rates (Foote, 1979). Taken together, these studies support the recommendation that a single mid-morning AI for all cows detected in estrus the night before or the same morning should result in near maximal conception rates and is an effective alternative to using the a.m./p.m. rule to manage AI to a detected estrus.

Table 4. Effect of time of AI relative to detection of estrus on 75 d nonreturn rates in lactating dairy cows (Nebel et al., 1994).

Interval (h)	Number of AI	75 d nonreturn rate (%)
0 - 6	1126	59.9[a]
6 - 12	2352	60.7[a]
12 - 18	2455	55.5[b]
18 - 24	962	53.4[b,c]
24 - 30	99	49.6[c]

[a,b,c]Means with different superscripts differ ($P < 0.01$)

Timing of AI to synchronized ovulations: Ovsynch56

A long standing goal of reproductive physiologists working with dairy cattle was to develop a hormonal synchronization program that could overcome the problems and limitations associated with visual detection of estrus. This goal was realized in 1995 with the publication of a hormonal synchronization protocol that combined GnRH and $PGF_{2\alpha}$ to control ovarian physiology and is now commonly referred to as the **Ovsynch** protocol (Pursley et al., 1995). The Ovsynch protocol synchronizes follicular development, luteal regression and ovulation such that artificial insemination can be conducted at a fixed-time without the need for estrus detection, commonly referred to as timed artificial insemination (**TAI**). Subsequent studies that repeated this work soon verified the results of the original publication (Burke et al., 1996; Pursley et al., 1997), and dairy producers and veterinarians began to implement the Ovsynch protocol as a tool for reproductive management on commercial dairies. Today many dairy farms in the U.S. and around the world have adopted systematic synchronization protocols as a management strategy for submitting cows for first and subsequent postpartum AI services (Caraviello et al., 2006).

Timing of the injections and artificial insemination in relation to the second GnRH of the Ovysnch protocol has been an area of recent scientific inquiry. The optimal timing of the second GnRH injection and TAI in an Ovsynch protocol was tested by Brusveen et al. (2008) by comparing two Cosynch protocols (i.e., Cosynch-48 and Cosynch-72 vs. Ovsynch-56. Lactating Holstein cows (n = 927 cows; n = 1,507 TAI) were blocked by pen on a commercial dairy, and pens were rotated among the three treatments. All cows received GnRH followed 7 d later by $PGF_{2\alpha}$ and then received one of the following treatments: 1) GnRH + timed AI 48 h after $PGF_{2\alpha}$ (Cosynch-48); 2) GnRH 56 h after $PGF_{2\alpha}$ + timed AI 72 h after $PGF_{2\alpha}$ (Ovsynch-56); or 3) GnRH + timed AI 72 h after $PGF_{2\alpha}$ (Cosynch-72). Overall fertility was similar for the Cosynch-48 (27%) and Cosynch-72 (23%) treatments, whereas cows receiving the Ovsynch-56 treatment had a greater fertility (36%) compared to Cosynch-48 or Cosynch-72 cows. A subsequent experiment conducted in 3 herds of lactating cows (n=739) confirmed the results of Brusveen et al. (2008). Cows receiving Cosynch-72 had lower fertility than cows receiving Ovsynch-56 (Nebel et al., 2008).

Table 5. One possible hormone injection and timed artificial insemination schedule for the Ovsynch-56 protocol based on the results of Brusveen et al. 2008.

Sunday	Monday	Tuesday	Wednesday	Thursday	Friday	Saturday
	GnRH - AM					
	PGF - AM		GnRH - PM	TAI - AM		

PGF = prostaglandin $F_{2\alpha}$; **GnRH** = gonadotropin-releasing hormone; **TAI** = timed artificial insemination; **AM** = ante meridiem; **PM** = post meridiem.

Based on results from these two experiments as well as an understanding of timing of AI in relation to ovulation, Ovsynch-56 (Table 5) is strongly recommended over Cosynch protocols which do not optimize the timing of AI in relation to ovulation. The Ovsynch portion of all subsequent protocols in this report should include the timing of the **Ovsynch-56** protocol for the second GnRH injection and TAI.

Key 3: Improve AI Efficiency – Who is breeding the cows?

AI efficiency refers to factors affecting pregnancy rates due to AI technique. Research has consistently shown that the people responsible for conducting AI on a farm can have a profound effect on fertility. A field trial was conducted to determine several factors regarding AI efficiency including whether conception rates achieved following AI by professional AI technicians and herdsman-inseminators differed (Dalton et al., 2004). Average conception rates of dairy cows was greater for professional AI technicians than for herdsman-inseminators (45 vs. 27%, respectively; Table 6), and the difference in mean conception rate achieved by professional AI technicians and herdsman-inseminators was not attributable to milk production, parity, service number or stage of lactation (Dalton et al., 2004). Interestingly, sequence of insemination did not affect conception rate for either category of inseminator indicating that four straws of semen can be thawed at a time as long as the time elapsed from thawing to insemination does not exceed 15 minutes (Dalton et al., 2004). These data strongly support use of professional AI technicians when AI technique is a suspected cause of a low conception risk on a farm.

Table 6. Effect of sequence of insemination on conception rates after simultaneous thawing of fourn 0.5-mL straws of semen by professional AI technicians and herdsmen-inseminators (Adapted from Dalton et al., 2004).

	Conception rate[1] (%)				
	Sequence of insemination				
Inseminator	1st straw	2nd straw	3rd straw	4th straw	Mean
AI technicians	40 (61/153)	47 (71/150)	41 (60/146)	50 (74/147)	45[a] (266/596)
Herdsmen	24 (26/108)	20 (21/103)	33 (36/110)	30 (33/108)	27[b] (116/429)

[a,b]Means with different superscripts differ (P<0.01).
[1]Within inseminator category, no differences were detected due to sequence of insemination.

Another example of the effect of AI technique on fertility was illustrated in a field trial in which nonlactating dairy heifers were inseminated by on-farm inseminators (Rivera et al., 2004). Nonlactating Holstein dairy heifers (n = 352) 13 mo of age were managed using a 42-d breeding period in which they were inseminated artificially (AI) after removed tail chalk evaluated once daily. Heifers were assigned randomly to either of two insemination schemes. At the onset of the breeding period (d 0), in one group of heifers ovulation was synchronized (100 µg GnRH, d 0; 25 mg $PGF_{2\alpha}$, d 6; 100 µg GnRH, d 8) before a timed artificial insemination (TAI; d 8); before and after TAI, inseminations were based on removed tail chalk for the entire AI breeding period (GPG; n = 175). A second group of heifers were inseminated during the entire AI breeding period (TC; n = 177) based solely on removed tail chalk. No treatment by AI technician interaction was detected (p=0.70); however, AI technician dramatically affected (P<0.01) conception rate (Table 7). Thus, the overall poor conception rates in this study can be attributed solely to differences due to poor AI efficiency of two of the inseminators. Clearly, the two suboptimal inseminators working for this custom heifer grower operation was limiting the reproductive success of the operation.

Table 7. Effect of inseminator on fertility of Holstein dairy heifers receiving AI after removed tail chalk (TC) or synchronization of ovulation and timed artificial insemination (GPG). (Adapted from Rivera et al., 2004).

	Inseminator		
Treatment	1	2	3
TC, %	30 (16/53)	33 (12/36)	63 (52/83)
GPG, %	20 (12/60)	25 (6/24)	54 (49/91)
Overall, %	25[a] (28/113)	30[a] (18/60)	58[b] (101/174)

[a,b]Within a row, percentages with different superscripts differ (P < 0.01). Treatment by inseminator interaction was not significant (P = 0.70).

Key 4: Identify nonpregnant cows early after an insemination (but not too early).

One technology that makes detection of nonpregnant cows early post breeding possible is transrectal ultrasonography, a technology that has been adopted by bovine practitioners in certain regions of the U.S. and around the world. Early pregnancy diagnosis improves reproductive performance by decreasing the interval between successive AI services and coupling a nonpregnancy diagnosis with an aggressive strategy to rapidly reinseminate these cows (Fricke, 2002). Although it has long been accepted that pregnancy status should be determined in dairy cows as soon as possible after AI, the accuracy of pregnancy diagnosis outcomes determined early after AI are confounded by subsequent pregnancy loss (Studer, 1969; Melrose, 1979). Recent research on the practical implementation of early pregnancy diagnosis using ultrasound in a systematic synchronization and resynchronization system has confirmed the notion that pregnancy diagnosis using ultrasound can be conducted too early and illustrates the pitfalls and limitations of early pregnancy diagnosis in lactating dairy cows (Fricke et al., 2003; Silva et al., 2007). Pregnancy loss diminishes the benefit of early pregnancy diagnosis in two ways. First, because of the high rate of pregnancy loss that occurs early, the magnitude of pregnancy loss detected is greater the earlier after TAI that a positive diagnosis is made. Thus, the earlier that pregnancy is diagnosed after TAI, the fewer nonpregnant cows are identified to which a management strategy can be implemented to resynchronize them. Second and more important, cows diagnosed pregnant earlier after TAI have a greater risk for subsequent pregnancy loss compared to cows diagnosed later after TAI. If left unidentified, cows diagnosed pregnant early after TAI that subsequently loose that pregnancy reduce reproductive efficiency by extending the interval from calving to the conception that results in a full-term pregnancy.

The accuracy of pregnancy outcomes using ultrasound after TAI was assessed in a field trial (Silva et al., 2007). Pregnancy examinations were performed by one herd veterinarian throughout the study using ultrasound in lactating Holstein cows (n=877) 27 d after first postpartum TAI. Outcomes were categorized as: pregnant (PG) = CL, normal uterine fluid, embryo visualized; questionable pregnant 1 (QP1) = CL, normal uterine fluid, embryo not visualized; questionable pregnant 2 (QP2) = CL, abnormal uterine fluid, embryo not visualized; pregnancy loss (PL) = nonviable embryo; nonpregnant (NP) = no CL and/or uterine fluid. Outcomes using US were compared to those categorized PG or NP using a pregnancy-associated glycoprotein (PAG) ELISA of plasma samples collected at the ultrasound diagnosis (Table 8). Outcomes for cows in which ultrasound and PAG outcomes agreed were considered correct, whereas cows in which outcomes disagreed were rechecked using ultrasound 32 d after TAI. These results demonstrate that although agreement between the PAG ELISA and ultrasound at 27 d after TAI was acceptable, ultrasound outcomes of QP1, QP2 and PL (23.9% of all US outcomes 27 d after TAI) were less accurate than PG or NP outcomes. Based on these results, early pregnancy diagnosis outcomes using ultrasound 27 d after TAI was less accurate than ultrasound 39 d after TAI.

Table 8. Frequency of pregnancy outcomes based on transrectal ultrasonography (TU) categories 27 d after timed AI and the frequency of incorrect TU outcomes based on pregnancy status re-evaluation using TU 32 d after timed AI (Adapted from Silva et al., 2007).

TU category	Frequency % (no./no.)	TU outcome disagreements with PAG ELISA % (no./no.)	Overall rate of incorrect TU outcomes % (no./no.)
PG	17.4 (295/1692)	5.1 (15/295)	2.4[a] (7/293)
QP1	19.6 (332/1692)	17.5 (58/332)	9.5[b] (31/325)
QP2	3.4 (58/1692)	65.5 (38/58)	57.4[c] (31/54)
Total pregnant	40.5 (685/1692)	16.2 (111/685)	10.3 (69/672)
PL	0.7 (11/1692)	36.4 (4/11)	18.2[b] (2/11)
NP	58.9 (996/1692)	8.5 (85/996)	2.0[a] (20/990)
Total not pregnant	59.5 (1007/1692)	8.8 (89/1007)	2.2 (22/1001)
Overall	100.0 (1692/1692)	11.8 (200/1692)	5.4 (91/1673)

[a,b,c] Within a column, values with different superscripts differ ($P < 0.05$).

Key 5: Aggressively re-inseminate nonpregnant cows.

Although use of synchronization of ovulation and TAI for improving service rate to first AI service reduces the impact of poor estrous detection, the improved AI submission rate to first TAI often is followed by a time lag exceeding 60 d before cows failing to conceive are detected and reinseminated. Because conception rates to TAI for dairy cows managed in confinement-based systems in the U.S. are reported to be 40% or less (Pursley et al., 1997; Fricke et al., 1998; Jobst et al., 2000), 60% or more of the cows will fail to conceive and therefore require a resynchronization strategy for aggressively initiating subsequent AI services. Methods for early detection of nonpregnancy coupled with hormonal resynchronization systems that program nonpregnant cows to receive subsequent TAI services are now being developed and assessed so that systematic reproductive management programs can be implemented to aggressively manage reproduction (Fricke, 2002). A wide array of reproductive management strategies have been developed to fit the various dairy production systems that are found around the world, and the diversity among these strategies has been reviewed elsewhere (Lucy et al., 2004).

One of the first field trials to directly compare intervals from first TAI to resynchronization of ovulation on a dairy incorporating transrectal ultrasonography for early pregnancy diagnosis was reported by Fricke et al. (2003). Lactating dairy cows (n=711) on a commercial dairy farm were enrolled into this study after a Presynch-Ovsynch protocol and were randomly assigned to each of three treatment groups for Resynch. All cows (n=235) in the first treatment (Day 19) received a GnRH injection 19 d after TAI and continued the Ovsynch protocol if diagnosed nonpregnant using US 26 d after TAI. Cows (n=240) in the second (Day 26) and cows (n=236) in the third (Day 33) treatments initiated Resynch if diagnosed not-pregnant using US 26 or 33 d after TAI, respectively. Overall P/AI to Resynch was 32% and was greater for D26 and D33 cows than for D19 cows (Fricke et al., 2003). Thus, the most aggressive Resynch interval tested in this experiment resulted in unacceptably poor fertility compared to delaying Resynch by 7 to 14 d.

Table 9. One possible hormone injection and timed artificial insemination schedule for the Resynch-32 protocol for second and greater TAI based on the results of Fricke et al. 2003.

Sunday	Monday	Tuesday	Wednesday	Thursday	Friday	Saturday
				TAI1		
	GnRH					
	PG+PGF		**GnRH**	**TAI2**		

PGF = prostaglandin $F_{2\alpha}$, **GnRH** = gonadotropin-releasing hormone, **TAI1**= first postpartum timed artificial insemination, **TAI2** = second and greater postpartum timed artificial insemination **PG** = pregnancy diagnosis

Although coupling a nonpregnancy diagnosis with a management strategy to quickly reinitiate AI may improve reproductive efficiency by decreasing the interval between AI services, early pregnancy loss and the effectiveness of Resynch initiated at certain physiologic stages post breeding may limit the effectiveness of the early Resynch strategies tested thus far. Thus, a justifiable Resynch strategy is a Resynch-32 protocol (Table 8) in which all cows are pretreated with GnRH 7 d before pregnancy diagnosis 32 days after TAI, identify cows failing to conceive to TAI and administer $PGF_{2\alpha}$ to cows diagnosed not-pregnant 39 days after TAI and complete the Resynch protocol (Table 9). This recommendation is based on data in which the earliest Resynch intervals of 19 or 26 d after TAI do not yield the greatest fertility (Fricke et al., 2003; Sterry et al., 2006) and the notion that assessment of pregnancy status should be delayed until the latest possible time after TAI and during Resynch to ensure that diagnostic outcomes using transrectal ultrasonography are not confounded by subsequent pregnancy loss (Silva et al., 2007). New resynch strategies are being tested to reduce the interval between TAI while also improving fertility and may displace the Resynch-32 protocol as the protocol of choice in the dairy industry.

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