Synchronization of Estrus and Ovulation in Dairy Cows

Jeffrey S. Stevenson

Department of Animal Sciences and Industry, Kansas State University, Manhattan, KS 66506-0201
Email: jstevens@oznet.ksu.edu

Take Home Message

- Management of the estrous cycle is now more practical than it was a decade ago because of our understanding of ovarian follicular waves.
- With availability of three gonadotropin-releasing hormone (GnRH) products and at least three prostaglandin products, the cycle can be controlled for timed artificial inseminations (TAI) with little loss in conception rate compared to inseminations made after detected estrus. Various systems are effective for programming first inseminations with or without some heat detection.
- With the incorporation of transrectal ultrasonography for early pregnancy diagnosis 28 to 30 days after insemination, routine heat detection programs could be eliminated by reprogramming each cow after an open diagnosis.
- The most limiting factor in the control of the cycle is the proportion of missed heats in estrus-synchronization programs that rely partly or solely on heat detection.
- Pregnancy rate (the proportion of cows that become pregnant of all cows programmed for insemination) is the best measure of an estrus-synchronization program because it measures total number of pregnancies achieved per unit of time rather than simple conception success at each insemination.

Introduction

Improving dairy herd reproductive management requires an understanding of the basic principles of getting cows pregnant. It is critical to understand each component of the estrous cycle as well as the annual reproductive cycle (calving interval) and determine where limited time and resources might be best concentrated to reach AI-breeding goals. A calving interval consists of four major components. The first component is the rest period or elective waiting period (EWP).

The second component is the period of time between the end of the EWP and when the first estrus is detected for the first AI-breeding. The duration of this period is a function of the heat detection rate as well as whether or not some hormonal regimen is used to bring cows into estrus after the end of the EWP.

The third component of a calving interval is the active AI-breeding period for each cow and represents the number of days required for the cow to conceive after the first AI service. If a cow conceives at first service, then the third component is nonexistent. Otherwise, it is a function of the heat detection rate and the level of herd fertility.

The fourth component of a calving interval is gestation. Based on these component parts of a calving interval, a EWP of 40 to 50 days is probably sufficient for essentially all cows. With a rate of heat detection of 65% and a conception rate of 65%, the average period from the end of the EWP until pregnancy is established in 95% of the cows should be 35 days. This means that some cows conceive immediately following the end of the EWP and others remain open for 100 or more days. With a EWP of 50 days, estrus and conception rates of 65%, and a gestation period of 280 days, an average calving interval of 365 days (50 + 35 + 280 = 365) is attainable, when it is desired that 95% of the cows conceive.

■ Programmed Breeding

Most dairy producers appreciate the benefits and advantages of using an estrus-synchronization program. Synchronizing estrus in cattle simply makes occurrence of estrus more predictable and AI-breeding more convenient. Dairy producers have benefited from the superior genetics of proven bulls, which have increased pride of ownership in better-bred cattle, as well as providing a payoff in greater milk production. Although most are sold on the idea of using heat synchronization, one question most frequently asked by dairy producers and dairy veterinarians is: What is the best way to synchronize estrus in dairy cows and heifers for AI-breeding?

The program used successfully on dairy farms is probably the one that is the most simple to execute. Although heat synchronization of large numbers of cows and heifers at one time is not typical on most dairy farms, except in large herds or where seasonal calving is practiced, one needs to develop a system for identifying cows (based on days after calving) and heifers (based on age) that should go into each breeding group cluster.

Breeding Clusters

The breeding cluster is one method that can be used to organize groups of cows for programmed breeding (Folman et al., 1984). For example, if the EWP is 50 d before scheduled AI, then a breeding cluster of cows can be organized to fall within a certain range of days in milk to fit the targeted first breeding date. These cows can be identified easily using DHIA software, computer records, or spreadsheet programs, or simply by keeping a chronological list of calving dates. In a herd of 200 cows, a cluster that calves during a 3-wk period can be organized so the freshest cow in the cluster meets the minimum acceptable EWP at the time of AI. When the EWP is 50 d, a cluster would consist of cows that are 50 to 70 d in milk during the targeted breeding week. Therefore, the average interval to first insemination for that cluster would be 60 days. Cows failing to conceive should return to estrus during the breeding week of the next cluster of cows, which would be estrus-synchronized for AI 3 wk after the first cluster. This clustering method allows first services and repeat inseminations to occur during the same week, thus concentrating most inseminations during 1 wk out of every 3 wk. This same system can be employed for AI of replacement heifers when they reach an acceptable age and weight to enter a breeding cluster.

In large herds (>200 cows), grouping cows into 2-wk clusters is recommended. These clusters simplify AI-breeding of cows that meet the breeding criteria on a biweekly basis. Therefore, during the period before the cows reach their targeted breeding date (based on days in milk and the EWP), estrus or ovulation is synchronized to occur during each breeding week. Usually, the synchronization period is set so estrus or TAI occurs during the Monday-to-Friday work week or at the convenience of scheduled labor.

Choosing a Breeding System

Once a system is in place to identify cows and heifers that fit those criteria for inclusion in an AI-breeding cluster, then the specific programmed breeding system is fit into a weekly management sequence. What successful programs are available? There are two general categories of programs from which to choose: 1) $PGF_{2\alpha}$; or 2) gonadotropin-releasing hormone (GnRH) + $PGF_{2\alpha}$ The first involves using either of two prostaglandin products that are available in the U.S. market (Lutalyse™ and Estrumate™). The second category uses either of three GnRH products (Cystorelin™, Fertagyl™, or Factrel™) plus a prostaglandin product in combination with heat detection or a TAI.

Targeted BreedingÔ Program. The Targeted Breeding program has been promoted by one of the $PGF_{2\alpha}$ manufacturers (Pharmacia & Upjohn) for synchronizing the AI-breeding of lactating cows in a herd (Figure 1; Nebel and

Jobst, 1998). Injections of $PGF_{2\alpha}$ are administered 14 days apart. This interval is simply based on the fact that sufficient time must pass after the first injection so those females responding to the first injection (their CL regresses and they come into estrus) have a new CL that is mature enough to respond to a second injection (at least on day 6 of the estrous cycle). In addition, those females that were not in a stage of the estrous cycle with a CL that could regress after the first $PGF_{2\alpha}$ injection should be responsive 14 days later. Targeted Breeding calls for the first injection (so-called set-up injection) to be given 14 days before the EWP ends. No cows are inseminated after the first injection, although up to 50% show estrus in response to the first injection. The second injection (first breeding injection) then is given just prior to the end of the EWP, so first services can occur when cows are eligible for AI-breeding. The Targeted Breeding program then suggests that if no estrus is detected after the second injection, a third injection (second breeding injection) is given in another 14 days. If no standing estrus is detected after this third injection, then one TAI can be given at 80 hr after this third injection of $PGF_{2\alpha}$.

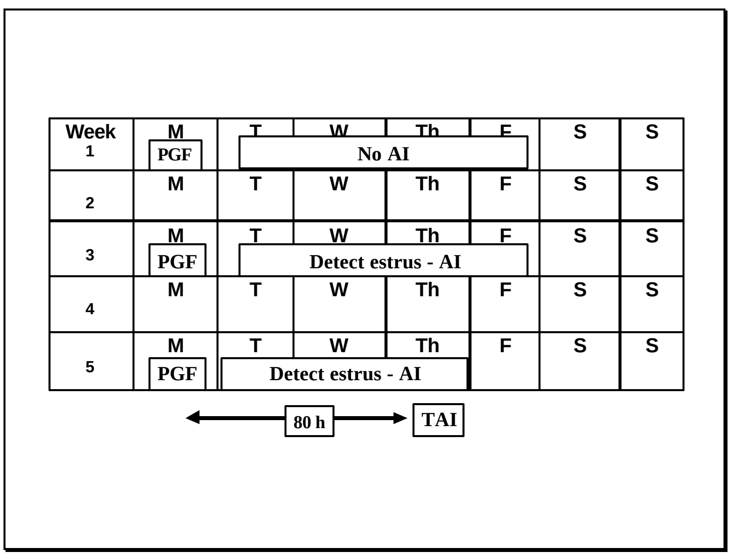

Figure 1. Injection schedule for the Targeted BreedingÔ program. Injections of PGF_{2a} are administered 14 d apart and inseminations occur after the second and third injections according to detected estrus. When no estrus is detected after the third injection, one timed AI (TAI) can be given 80 h later.

Ovsynch. The second method (named Ovsynch; Pursley et al., 1997a,b) is similar to the previous program, except it requires no heat detection (Figure 2). In fact, it is described more accurately as an ovulation synchronization program; hence the name, Ovsynch. A 100-µg injection of GnRH is given 7 days before a $PGF_{2\alpha}$ injection, and then a second 100-µg injection of GnRH is administered 36 to 48 hr after $PGF_{2\alpha}$, with one TAI given 0 to 24 hr later. A recent study found that 1 mL or 50 µg of Cystorelin is sufficient (Fricke et al., 1998). The first GnRH injection alters follicular growth by inducing ovulation of the largest follicle (dominant follicle) in the ovaries after the GnRH injection to form a new or additional CL (Pursley et al., 1995). Thus, estrus usually does not occur until after a PGF_2 injection regresses the natural CL and the secondary CL (formed from the follicle induced to ovulate by the first GnRH injection). Therefore, a new group of follicles appears in the ovaries (based on transrectal ultrasonographic evidence) within 1 to 2 days after the first injection of GnRH (Vasconcelos et al., 1999). From that new group of follicles, a newly developed dominant follicle emerges, matures, and can ovulate after estrus is induced by $PGF_{2\alpha}$ or it can be induced to ovulate after a second injection of GnRH. The GnRH injections release pituitary luteinizing hormone (LH), the natural ovulation-inducing hormone of the estrous cycle. Few cows will show heat in this program (Stevenson et al., 1999). About 8 to 16% may show heat around the time of the $PGF_{2\alpha}$ injection. If so, those cows should be AI-bred according to the AM-PM rule and the second GnRH injection eliminated.

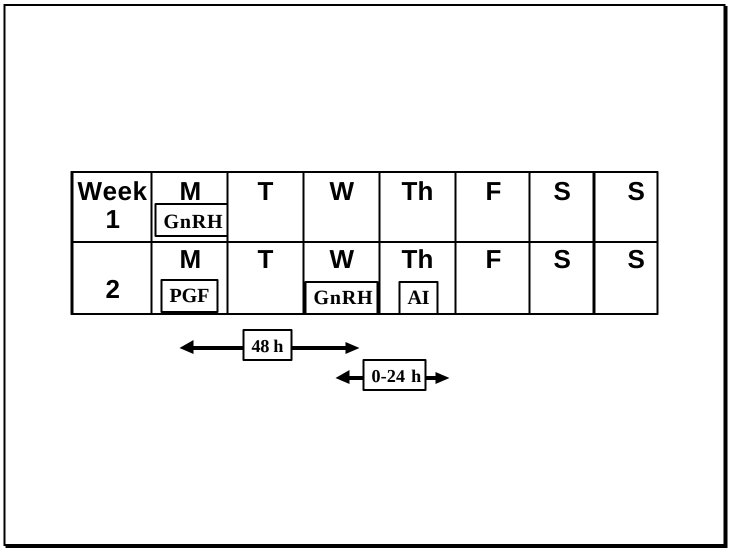

Figure 2. Injection schedule for the OvsynchÔ protocol. Injections of gonadotropin-releasing hormone (GnRH) are administered 7 d before and 48 h after $PGF_{2\alpha}$. Cows are inseminated between 0 and 24 h after the second GnRH injection.

This program works in replacement heifers, but because of lower pregnancy rates than can be achieved with other programs (Stevenson et al., 2000), it generally is not recommended unless detection of estrus during 40 hr after $PGF_{2\alpha}$ and subsequent AI are combined with TAI for all heifers not in estrus by 40 hr followed immediately by a second injection of GnRH (Schmitt et al., 1996). For some unexplained reason, the first GnRH injection fails to result in ovulation of a follicle as often in heifers as in lactating cows (Pursley et al., 1995).

When TAI is performed in cows that you are attempting to AI-breed, then by definition conception rate (CR) is the same as pregnancy rate (PR), because the heat detection rate (HDR or AI submission rate) is 100%. Therefore, PR = HDR $\times$ CR becomes PR = 1 $\times$ CR or PR = CR. For example, let's compare a traditional AI program that uses heat detection to Ovsynch in which no heat detection is necessary prior to first service (Table 1). If 70% of the cows in the traditional program are submitted for insemination (70% heat detection rate), with a 50% conception rate, 35% of the cows become pregnant in a 21-day period. With an Ovsynch program, 100% of the cows are inseminated, and with a similar conception rate, 50% of the cows become pregnant in a 10-day period. Therefore, 15 more pregnancies are achieved at a similar conception rate because all cows eligible for insemination are AI-bred; or in other words, 30 eligible cows in the traditional program were not inseminated because they were not detected in heat. Therefore, more pregnancies can be established per unit of time.

Table 1. Pregnancy Rates Achieved with Traditional Heat Detection[1] and Ovsynch[2] Programs

Item	Traditional	Ovsynch
No. of cows attempted for AI in 21 days	100	100
No. of cows submitted for AI (heat detection rate; HDR), %	70	100
Conception rate (CR), %	50	50
Pregnancy rate (PR)[3], %	35	50

[1]Observation for estrus and no hormone use or estrus-synchronization program.

[2]See Figure 2.

[3]PR = HDR $\times$ CR.

Presynch + Ovsynch. Further research with the Ovsynch protocol revealed that when cows were started on this protocol between days 5 and 12 of the estrous cycle, conception rates tended to be greater than at other stages of the cycle (Figure 3; Vasconcelos et al., 1999). As a result of those findings, at least three experiments have demonstrated improved fertility in cows when one $PGF_{2\alpha}$ injection was given 12 days before starting the Ovsynch protocol or two injections were given 14 days apart, with the second injection given 12 days before starting the Ovsynch protocol. In those studies, pregnancy rates of multiparous cows were increased by 13 percentage points with one injection of $PGF_{2\alpha}$ preceding the Ovsynch protocol, (Cartmill et al., 2000a), whereas pregnancies were improved by 12 to 14 percentage points in all lactating cows with two injections of $PGF_{2\alpha}$ (Moreira et al., 2000; El-Zarkouny and Stevenson, unpublished results). The Presynch procedure entails two injections of $PGF_{2\alpha}$, given 14 days apart with the second injection given 12 days before initiating the Ovsynch protocol (Figure 3).

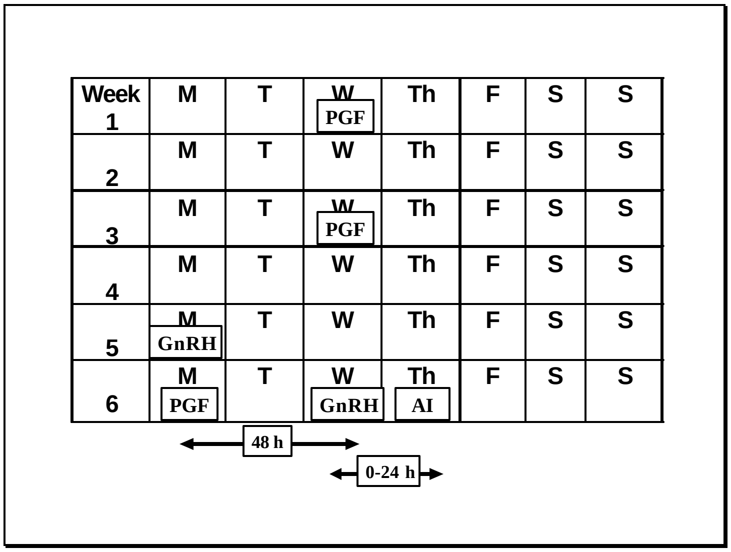

Figure 3. Injection schedule for the Presynch protocol that precedes the Ovsynch protocol. This protocol is similar to the Ovsynch protocol except that two injections of $PGF_{2\alpha}$ are administered 14 d apart with the second given 12 d before initiating the Ovsynch protocol. The purpose of the $PGF_{2\alpha}$ injections is to group cows into the early luteal phase of the estrous cycle at the onset of the Ovsynch protocol.

▪ Cows Not Pregnant at Pregnancy Diagnosis

A traditional procedure has been to give a $PGF_{2\alpha}$ injection to any cow with a palpable CL diagnosed open at pregnancy checks. Cows were either inseminated based on subsequently detected estrus or inseminated once (80 hr; Elmarimi et al., 1983) or twice (72 and 96 hr; Plunkett et al., 1984) after $PGF_{2\alpha}$. Because inseminations are based only on detected estrus, some open cows may go without reinsemination for weeks. The Ovsynch program is ideal for preparing open cows for reinsemination (Figure 4). It guarantees that every cow is reinseminated within at least 10 days after nonpregnancy status is identified. If the open cow is detected in estrus at any time during the 10-d protocol, then the cow should be inseminated according to detected estrus and the remaining portions of the protocol discontinued. Using this system, detection of estrus can be eliminated totally, but inter-insemination intervals will average 50 days (time to pregnancy diagnosis + 10 d for the Ovsynch protocol) rather than multiples of one estrous cycle (approximately 21 days) depending on the rate of detected estrus in the herd.

Week	MON	TUE	WED	THU	FRI	SAT	SUN
1	GnRH						
2	PGF		GnRH	AI			
	40 d						
8	Pregnancy diagnosis: Open? GnRH						
9	PGF		GnRH	AI			

Figure 4. Programmed reproductive management without detection of estrus can occur when using the Ovsynch or Presynch+Ovsynch protocols. When cows are diagnosed open based on weekly Monday morning palpation, all nonpregnant cows are restarted on the Ovsynch protocol and thus time-inseminated 10 d after their nonpregnancy status is identified.

■ Costs of Programmed AI-Breeding

Assessing the costs of using programmed AI-breeding is not easy. Further, most producers assume that it is more costly because of the extra labor, semen, and hormones. Table 2 summarizes how programmed breeding pays for itself. Let's assume that you are using Ovsynch and want to compare that to AI-breeding cows based on heat detection, perhaps coupled with tail chalk, tail paint, or even Kamar™ or Bovine Beacon™ heat-mount patches. The total cost of Ovsynch is about $38 ($13 for the three injections, $5 for labor to administer injections, $15 for semen, and $5 for AI-breeding). That compares to $20 (semen + AI-breeding) for the traditional approach. If we assume that conception rate is 40% in both cases, then at a 70% heat detection rate, the traditional program would produce 28 pregnancies (70 × 40) and Ovsynch would produce 12 more pregnancies or 40 in total.

Table 2. Comparison of AI-Breeding Costs of Ovsynch and a Traditional Heat Detection Program without Hormonal Intervention

Per Cow	Traditional		Ovsynch	
Hormones[1], $	0		13	
Labor, $	0		5	
Semen + AI[2], $	20		20	
Total costs, $	20		38	
Per 100 Cows	Heat detection rate		AI submission rate	
No. of cows inseminated	50	70	100	100
No. of pregnancies[3]	20	28	40	40
Cost for 100 cows[4], $	1000	1400	3800	3800
Cost per pregnancy[5], $	50	50	95	95
More pregnancies by Ovsynch[6]			+20	+12
Total cost of additional pregnancies[7], $			2800	2400
Per cow cost of additional pregnancies[8], $			140	200
Value of additional pregnancy[9], $			274	253
Semen + AI labor, $			50	50
Additional days open at $1 per day			84	63
Replacement cost, $			140	140
Net return per additional pregnancy, $			+134	+53

Source: Adapted from Hoard's Dairyman, September 10, 1998, p. 662.

[1] Cost of PGF_2 = $3 and two doses of GnRH = $5.

[2] Cost of semen = $15 and insemination = $5.

[3] No. of pregnancies or pregnancy rate = heat detection rate × conception rate (40%).

[4] No. inseminated (50, 70, or 100) × cost per cow.

[5] Cost per 100 cows divided by the number of pregnancies.

[6] Compared to 50% and 70% heat detection rates, respectively.

[7] Difference in cost for the traditional and Ovsynch programs at each heat detection level.

[8] Cost of additional pregnancies divided by the number of pregnancies.

[9] Cost of 2.5 more services (40% conception rate) + average of 63 or 84 days open to impregnate successfully 80% of the 12 or 20 remaining open cows (not pregnant after first service in the traditional program), respectively, + the cost of replacing 20% of open cows with replacements valued at $1200 each and cull cows worth $500.

What is the additional value of those 12 pregnancies? To determine this, we need to estimate the value of one pregnancy after the cow has already failed to conceive once. It takes about 63 days to get a cow pregnant after the first unsuccessful service, so at only $1 per day, the pregnant cow has a $63 greater value compared to the nonpregnant cow. On average 2.5 more doses of semen + AI labor will be needed or $50 more per pregnancy. If we assume that 20% of the cows will fail to conceive, the cost of a replacement heifer is $1200, and the value of a cull cow is $500, then we must add $140 ($700 × 20% culls). So one additional pregnancy is worth $253 ($63 + $50 + $140). Because those 12 additional pregnancies cost us $200 each, we have a positive return on our investment of $53 per additional pregnancy.

Now if heat detection is closer to 50% as in most herds, then only 20 pregnancies are achieved in 21 days and that is 20 less that what is achieved with Ovsynch. Each of those pregnancies would cost only $140 ($3800 Ovsynch costs - $1000 traditional costs/20). Because of poorer heat detection, it will take one more estrous cycle or 84 days to get 80% of the remaining cows pregnant, so the value of a pregnant cow is $84 more than that of the open cow. The costs of semen, AI-breeding, and culling are the same, so the value of one additional pregnancy at a 50%-heat detection rate is $274 ($84 + $50 + $140). That means the cost of $140 per each additional pregnancy gained by Ovsynch gives a positive return of $134 per additional pregnancy. Clearly, Ovsynch or other programmed AI-breeding systems can pay for themselves because more cows become pregnant per unit of time, so even though more costs are associated with their use, the return on investment is greater. Based on these cost estimates, as heat detection, conception rates, or both decline, the programmed AI-breeding, in this case, Ovsynch, pays for itself.

These differences between the two programs might be even greater, if the costs of heat detection and tail chalk, tail paint, or heat-mount detectors in the traditional program were included. We know that heat detection cannot be eliminated completely, so it leaves us wondering how to estimate the real costs of administering a programmed breeding system. Of course, many variables determine the cost-benefit ratio of a given system on each farm, for example, the number of cows, type of housing, cost and availability of skilled labor. The selection of the best programed-breeding system for an individual herd also depends on that herd's rate of heat detection. Those herds with excellent heat expression and(or) heat detection may be served best by programs with less hormonal intervention.

▪ Advantages of Various Programs

Table 3 summarizes the advantages of the three programmed breeding systems described above. The best program is the simplest and easiest to manage, and that may vary with personalities and skills of personnel for each herd. Simplicity could be defined as the fewest number of cow handlings per program. Some studies show that conception rates are greater when cows are inseminated after some natural estrus compared to a TAI, whereas others detected no differences (Stevenson et al., 1996; Pursley et al., 1997a,b; Cartmill et al., 2001b). Few comparisons of programmed-breeding systems have been made in field trials (Nebel and Jobst, 1998). Pregnancy rates achieved in several herds after the Targeted Breeding and Ovsynch programs were similar for lactating cows but less for replacement heifers on the latter program. Both programs improved reproductive performance over traditional programs (Nebel and Jobst, 1998) and decreased days open compared to no hormonal intervention.

Table 3. Advantages of various programmed-breeding systems

	Programs		
Item	TB[1]	Ovsynch	Presynch + Ovsynch
Simplicity		X	
Greatest conception rates[2]	X		
Greatest pregnancy rates[3]		X	X
Herd size	X	X	X
No detection of estrus		X	X
Shorter time of program administration		X	
Synchronizes follicle growth and CL regression		X	X
Timed insemination without detection of estrus		X	X

[1]TB = Targeted Breeding

[2]Conception rate = no. of pregnant cows divided by the number of cows inseminated.

[3]Pregnancy rate = no. of pregnant cows divided by the number of cows treated. When timed AI is used and the AI-submission rate is 100%, then conception rate = pregnancy rate.

The important point is not the comparison of conception rates, but whether one program produces more pregnancies per unit of time (pregnancy rate) compared with another. If conception rates are similar, pregnancy rates will be greater when TAI is used because the AI submission rates are 100%.

Therefore, all programs described except the Targeted Breeding program have the TAI component. That program requires considerably more time to administer, including the need for detection of estrus. Where skilled cow people are involved and are adept in detecting subtle as well as major symptoms of estrus, the Targeted Breeding program may be successful. The disadvantage of any system is the rigidity of the injection schedules to which one must conform to guarantee its success.

What adds to the complexity of these systems is the overlapping nature of the cluster groups. For example, if the Presynch + Ovsynch protocol is followed where a new cluster is initiated every 2 wk, injections must be given concurrently to three different cluster groups. Further, if pregnancy is diagnosed once weekly and open cows are started in their own separate cluster group, the complexity of more cluster groups increases. In almost every case, personnel must be disciplined and detailed in their approach to making these programs work. Otherwise, the alternative is few pregnancies achieved and high culling rates of open cows. As herd size increases and more cows are confined to concrete, the efficiency of detected estrus will decline, because the number of standing events and duration of estrus are less on concrete where footing is not as good as it is on dirt (Britt et al., 1986). As a result of less inefficient detection of estrus or the lack of expressed estrus by cows in environments with poor footing, TAI programs will become more popular because of their predictability and manageability and likely produce a greater proportion of the total pregnancies of dairy cows in the future.

▪ References

Britt, J. H., R. G. Scott, J. D. Armstrong, and M. D. Whitacre. (1986) Determinants of estrous behavior in lactating Holstein cows. J. Dairy. Sci. 69:2195-2202.

Cartmill, J. A., S. Z. El-Zarkouny, B. A. Hensley, G. C. Lamb, and J. S. Stevenson. (2001a) Stage of cycle, incidence and timing of ovulation, and pregnancy rates in dairy cattle after three timed breeding protocols. J. Dairy Sci. 84:In press.

Cartmill, J. A., B. A. Hensley, S. Z. El-Zarkouny, T. G. Rozell, J. F. Smith, and J. S. Stevenson. (2001b). An alternative AI-breeding protocol for dairy cows exposed to elevated ambient temperatures before or after calving or both. J. Dairy Sci. 84:In press.

Elmarimi, A. A., C. D. Gibson, D. Morrow, J. Marteniuk, and B. Gerloff. (1983) Use of prostaglandin F2alpha in the treatment of unobserved estrus in lactating dairy cattle. Am. J. Vet. Res. 44: 1081-1084.

Folman, Y., M. Kaim, Z. Herz, and M. Rosenberg. (1984) Reproductive management of dairy cattle based upon synchronization of estrous cycles. J. Dairy Sci. 67:153-160.

Fricke, P.M., J. N. Guenther, and M. C. Wiltbank. (1998) Efficacy of decreasing the dose of GnRH used in a protocol for synchronization of ovulation and timed AI in lactating dairy cows. Theriogenology 50:1275-1284.

Moreira, F., C. A. Risco, M.F.A. Pires, J. D. Ambrose, M. Drost, and W. W. Thatcher. (2000) Use of bovine somatotropin in lactating dairy cows receiving timed artificial insemination. J. Dairy Sci. 83:1237-1247.

Nebel, R. L., and S. M. Jobst. (1998) Evaluation of systematic breeding programs for lactating dairy cows: a review. J. Dairy Sci. 81:1169-1174.

Plunkett, S. S., E. P. Call, and J. S. Stevenson. (1984) Prostaglandin $F_{2\alpha}$ for lactating dairy cows with a palpable corpus luteum but unobserved estrus. J. Dairy Sci. 67:380-387.

Pursley, J. R., M. R. Kosorok, and M. C. Wiltbank. (1997a) 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 , and GnRH. Theriogenology 44:915-923.

Pursley, J. R., M. C. Wiltbank, J. S. Stevenson, J. S. Ottobre, H. A. Garverick, and L. L. Anderson. (1997b) Pregnancy rates per artificial insemination for cows and heifers inseminated at a synchronized ovulation or synchronized estrus. J. Dairy Sci. 80:295-300.

Schmitt, E. J., T. Diaz, M. Drost, and W. W. Thatcher. (1996) Use of a gonadotropin-releasing hormone agonist or human chorionic gonadotropin for timed insemination in cattle. J. Anim. Sci. 74:1084-1091.

Stevenson, J. S., Y. Kobayashi, M. P. Shipka, and K. C. Rauchholz. (1996). Altering conception of dairy cattle by gonadotropin-releasing hormone preceding luteolysis induced by prostaglandin F_2 . J. Dairy Sci. 79:402-410.

Stevenson, J. S., Y. Kobayashi, and K. E. Thompson. (1999) Reproductive performance of dairy cows in various programmed breeding systems including Ovsynch and combinations of gonadotropin-releasing hormone and prostaglandin F_2 . J. Dairy Sci. 82:506-515.

Stevenson, J. S., J. F. Smith, and D. E. Hawkins. (2000) Reproductive outcomes of dairy heifers treated with combinations of prostaglandin F_2 , norgestomet, and gonadotropin-releasing hormone. J. Dairy Sci. 83:1-8.

Vasconcelos, J.L.M., R. W. Silcox, G. J. Rosa, J. R. Pursley, and M. C. Wiltbank. (1999) Synchronization rate, size of the ovulatory follicle, and pregnancy rate after synchronization of ovulation beginning on different days of the estrous cycle in lactating dairy cows. Theriogenology 52:1067-1078.