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## Application of the progesterone (CIDR) insert in artificial insemination programs of dairy cattle

### Abstract

Use of progesterone inserts (controlled interval drug release, CIDR) offers another option for synchronizing estrus and ovulation in replacement heifers and lactating dairy cows. Results indicate that heifers may be inseminated after detected estrus, at a fixed time (timed AI), or a combination of both. Conception rates exceed 50% in both scenarios. Practical applications of the CIDR in lactating cows have been used to resynchronize the return estrus of previously inseminated cows and as part of first-service AI-breeding protocols. Use for resynchronization has no drawbacks in previously inseminated cows, but may increase embryo survival during the first 30- to 60-days of pregnancy. No increase in the heat-detection rates of open cows is generally achieved, and no differences in return conception rates are observed between treated and control cows. First-service applications of the progesterone insert have resulted in some positive and some negative effects on timed AI (TAI) conception rates. More research is warranted to determine if an identifiable subpopulation of cows can benefit from exposure of the progesterone insert before first AI.; Dairy Day, 2007, Kansas State University, Manhattan, KS, 2007; Dairy Research, 2007 is known as Dairy Day, 2007

### Keywords

Dairy Day, 2007; Kansas Agricultural Experiment Station contribution; no. 08-127-S; Report of progress (Kansas State University. Agricultural Experiment Station and Cooperative Extension Service); 984; Dairy; Heifers; Cows; CIDR; Ovsynch; Pregnancy rates

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## **APPLICATION OF THE PROGESTERONE (CIDR) INSERT IN ARTIFICIAL INSEMINATION PROGRAMS OF DAIRY CATTLE**

*J. S. Stevenson*

### **Summary**

Use of progesterone inserts (controlled interval drug release, CIDR) offers another option for synchronizing estrus and ovulation in replacement heifers and lactating dairy cows. Results indicate that heifers may be inseminated after detected estrus, at a fixed time (timed AI), or a combination of both. Conception rates exceed 50% in both scenarios. Practical applications of the CIDR in lactating cows have been used to resynchronize the return estrus of previously inseminated cows and as part of first-service AI-breeding protocols. Use for resynchronization has no drawbacks in previously inseminated cows, but may increase embryo survival during the first 30- to 60-days of pregnancy. No increase in the heat-detection rates of open cows is generally achieved, and no differences in return conception rates are observed between treated and control cows. First-service applications of the progesterone insert have resulted in some positive and some negative effects on timed AI (TAI) conception rates. More research is warranted to determine if an identifiable subpopulation of cows can benefit from exposure of the progesterone insert before first AI.

(Key words: heifers, cows, CIDR, Ovsynch, pregnancy rates.)

### **Introduction**

Because fertility of lactating dairy cows is poor and has decreased more than 50% since 1970, improving fertility of lactating dairy cows is economically important to dairy pro-

ducers. A negative relationship exists between dry matter intake and circulating concentrations of progesterone in lactating dairy cows. Lactating dairy cows have serum concentrations of progesterone less than those in heifers. Progesterone is important to fertility, as demonstrated by a positive correlation between serum progesterone before AI and subsequent conception rate. Progesterone, in addition to use of gonadotropin releasing hormone (GnRH) and prostaglandin F<sub>2α</sub> (PGF<sub>2α</sub>), can be used to facilitate AI by synchronizing estrus and ovulation in heifers and lactating cows. Understanding the pharmacological impact of progesterone supplementation on conception rates may lead to a better understanding of the physiological reasons for reduced fertility of lactating dairy cows.

Today, progesterone is applied in the form of an intravaginally placed insert containing 1.38 g of progesterone and is marketed as an EAZI-BREED CIDR. This is the only progesterone product approved by the U.S. Food and Drug Administration for use in cattle. Its label indications include synchronization of estrus in suckled beef cows and replacement beef and dairy heifers, advancement of first postpartum estrus in suckled beef cows, and advancement of first pubertal estrus in beef heifers. For these applications, the insert is left in place for 7 days, with an injection of Lutalyse 1 day before insert removal. Signs of estrus are then observed on days 1 to 3 after insert removal, and insemination occurs about 12 hours after the first signs of estrus.

Label indications for lactating dairy cows include synchronization of the return estrus for cows previously inseminated. The insert is applied  $14 \pm 1$  days after insemination and the insert is removed 7 days later. Again, signs of estrus are observed on days 1 to 3 after insert removal, and insemination should occur about 12 hours after detection of estrus.

The objective of this review is to provide a summary of several proven applications of progesterone in successful AI-breeding programs. Most of the programs discussed are considered to be extra label, however, their application goal is to synchronize estrus and ovulation before insemination, either based on signs of estrus or by appointment (fixed-time AI or TAI).

### **Replacement Heifers**

Injecting  $\text{PGF}_{2\alpha}$  at the end of a short-duration (7 days) CIDR treatment allows estrus in nearly all females to be synchronized during a shorter treatment period. Administering progesterone for approximately 7 days allows any female in estrus or metestrus at the outset to advance to the luteal phase before  $\text{PGF}_{2\alpha}$  is administered. Expression of estrus is prevented in the remaining females that are in proestrus or late diestrus by the progesterone insert, and those in the luteal phase respond to the injection of  $\text{PGF}_{2\alpha}$ . Therefore, the combination of both hormones shortens the overall period of treatment and generally allows normal fertility.

The standard labeled indication protocol is illustrated in Figure 1 (Option A) in which heifers are inseminated after detection of estrus. Average occurrence of estrus is about  $43 \pm 3$  hours after insert removal, if  $\text{PGF}_{2\alpha}$  is administered 24 hours before insert removal, or  $49 \pm 1$  hours if the injection occurs at insert removal. Conception and pregnancy rates are shown for Options B and C. In Option B, heif-

ers were treated at 3 locations in Kansas. In Option C, heifers were treated at 12 locations in the 6 Midwest states. Combining heat detection to 84 hours after insert removal and a cleanup TAI at 84 hours resulted in 85% of the inseminations made before the cleanup TAI. The latter two treatments administered one TAI at 60 hours. Note that measures of fertility are quite similar and administering GnRH upfront resulted in very little increase in fertility. The most consistent results at all 12 locations in Option C, however, was the last treatment, in which GnRH was administered upfront with the insert and one TAI was administered at 60 hours after insert removal.

### **Lactating Dairy Cows**

#### ***Synchronizing Return to Estrus***

Three studies have been conducted in which the CIDR insert was used to resynchronize return to estrus in lactating dairy cows (Table 1). Study 1 was conducted in 2 Kansas herds. The CIDR was inserted for 7 days beginning at day 13 after TAI (first service, Figure 2). Cows were observed for signs of estrus and re-inseminated between 20 and 26 days after TAI. Study 2 was conducted in 7 herds located in 5 states. These cows were treated with a single injection of Lutalyse and inseminated. Beginning  $14 \pm$  days after AI, the progesterone insert was placed intravaginally for 7 days. Cows were observed for estrus and inseminated between 21 and 25 days after AI. Study 3 was conducted in California in 1 commercial dairy herd. Treatment with the CIDR insert occurred at 14 days after TAI (first service).

Results of the 3 studies are found in Table 1. In 1 of 3 studies, conception rate of cows that became pregnant before treatment was slightly reduced, but in 2 studies, pregnancy loss of cows that became pregnant before treatment was reduced in response to the progesterone treatment during the third week of

pregnancy ( $14 \pm 1$  to  $21 \pm 1$  days). Heat-detection return rates were increased in study 2; but overall, the percentage of cows returning to estrus for re-insemination at the first eligible estrus did not differ from controls. Conception rate of the returned estrus was reduced in study 1, but overall, no differences were detected between treatments.

These studies clearly show that no harm is done to cows of unknown pregnancy status when treated with the progesterone insert  $14 \pm 1$  days after insemination. Return heat-detection rates are neither improved, nor are return conception rates reduced. Embryo survival seemed to be improved in pregnant cows treated with the progesterone insert. These studies illustrate that 34 to 38% of cows return to estrus after  $21 \pm 1$  days from previous AI. Emphasis on heat detection during this period is important to reducing inter-insemination intervals in cows.

### ***Use Before First Services***

Seven studies have been conducted in which the progesterone insert was incorporated into the Ovsynch or Presynch + Ovsynch protocols for lactating dairy cows receiving their first AI after calving (Figure 2). Conception rates are reported for each of these studies in Table 2. In 4 studies in which the comparison was Ovsynch vs. Ovsynch + CIDR, 3 studies (studies 1, 3, and 6) reported greater TAI conception rates in cows receiving progesterone. Conception rates, however, for older cows of study 3 and for all cows in study 2, to which the CIDR insert was applied, were less than those in nontreated cows.

In 4 studies in which the comparison was Presynch + Ovsynch vs. Presynch + Ovsynch + CIDR, positive results were reported in 2 studies (studies 4 and 7; Table 2). For cows treated with the progesterone insert (studies 2 and 5), TAI conception rates were less than in cows not treated. These obvious inconsisten-

cies are further compounded by the fact that in studies in which multiple herds were treated (e.g., study 6), a treatment  $\times$  herd interaction was detected. The interaction indicates that the treatment (addition of progesterone) was only positive in some herds.

Closer examination of 2 of the studies cited previously (studies 6 and 7), indicates that progesterone is beneficial in a subpopulation of all cows treated (Table 3). Based on blood samples collected before treatments were applied, the cycling status of cows in these 2 studies was determined. When cows had been exposed to elevated concentrations of progesterone ( $\geq 1$  ng/mL) in either 2 or 3 samples collected during 10 or 28 days before treatment, cows were classified as cycling, whereas those having consistently low ( $< 1$  ng/mL) concentrations during the same period were classified as noncycling. Table 3 illustrates these 2 classifications of cows, plus whether they had elevated concentrations of progesterone when the progesterone insert was removed and PGF<sub>2 $\alpha$</sub>  was injected before TAI.

Noncycling cows having elevated progesterone at insert removal in study 1 were known to have functional luteal tissue, because the insert was removed at least 1 hour before blood was collected to measure progesterone. In study 2, blood was collected at insert removal. For noncycling cows in study 1 with elevated progesterone before PGF<sub>2 $\alpha$</sub>  was injected, a follicle must have ovulated in response to the first GnRH injection of the Ovsynch protocol and formed a corpus luteum (CL). In these cows, the CIDR insert seemed to provide no benefit (Table 3). In contrast, noncycling cows having low progesterone before PGF<sub>2 $\alpha$</sub>  was injected remained anovulatory or noncycling and may have benefited from exposure to the insert in study 1, because TAI conception rates were three-fold greater than in no-CIDR cows (Table 3). Caution in interpreting a benefit is suggested because of the

small numbers of cows in that classification (noncycling + low concentrations). Further, this benefit was not observed for similarly treated cows in study 2, nor in another study in which TAI conception rates were 18% for 38 CIDR-treated cows and 19% for 32 nontreated cows.

Cycling cows having elevated concentrations of progesterone before PGF<sub>2α</sub> was injected were those having a functional CL at insert removal, whereas those having low concentrations at insert removal had no functional CL (likely regressed during progesterone treatment). Cycling cows, particularly those cows in which no functional luteal tissue was present at insert removal, exposed to the progesterone insert during 7 days before PGF<sub>2α</sub> was injected had greater TAI conception rates. In this case, the progesterone insert prevented premature estrus and ovulation. For comparison purposes, study 2 included cows having a CL at the onset of the Ovsynch protocol and not treated with a CIDR insert. Their TAI conception rates are shown in Table 3.

At present, it is difficult to make any recommendation about how the CIDR insert might be used to improve TAI conception rates in lactating dairy cows. Because of its cost (approximately \$10), the insert should not be used without sound evidence for its benefit

to fertility and a return on its investment to dairy producers.

One recently published study applied a “cherry picking” approach to use of the CIDR insert. This study was conducted in a dry lot dairy. Cows were treated with the Presynch + Ovsynch protocol. Presynch PGF<sub>2α</sub> injections were administered at 47 and 61 days in milk and cows detected in estrus were inseminated after *either* Presynch injection. Heat detection allowed 77% (3,974 of 5,162) of the cows to be inseminated during 33 days that followed the 2 Presynch injections until Ovsynch (n = 589) or Ovsynch + CIDR (n = 586) was applied to the remaining noninseminated cows. Those remaining cows exposed to the insert had greater TAI conception rates than nontreated cows (31 vs. 23%). Further, cows exposed to progesterone had greater concentrations of progesterone 14 days after TAI than nontreated cows, regardless of pregnancy status. Results of this study indicate that a population of cows may be responsive to the benefit of progesterone treatment. This population of cows may represent a larger proportion of noncycling cows and those which poorly express estrus. Another large-scale multi-herd study is currently being conducted to verify the results of the previous study.

**Table 1. Responses of Lactating Cows to CIDR Insert to Synchronize Return of Estrus**

Study	Treatment	Previous AI			Return AI		
		No. of cows	Conception rate, %	Pregnancy loss, %	No. of cows	Return rate, %	Conception rate, %
1	CIDR	297	42	34*	169	31	20*
	Control	327	38	56	189	27	32
2	CIDR	881	33*	...	589	34*	27
	Control	863	37	...	544	19	31
3	CIDR	373	36	16*	227	55	31
	Control	602	32	25	387	59	27
Total	CIDR	1,551	35	25**	985	38	27
	Control	1,792	36	33	1,120	34	30

\*Differs ( $P < 0.05$ ) from control within study.

\*\*Differs ( $P < 0.05$ ) from control across all studies.

**Table 2. Synchronization of Ovulation in Lactating Dairy Cows after Ovsynch or Presynch + Ovsynch Compared with Similar Treatments that Included a Progesterone Insert (CIDR)**

Study	Days since timed AI	Ovsynch		Presynch + Ovsynch	
		No CIDR	CIDR	No CIDR	CIDR
----- No./no. (%) -----					
1	29 <sup>1</sup>	33/91 (36)	54/91 (59*)		
	57 <sup>1</sup>	18/91 (20)	41/91 (45*)		
2	29 <sup>1</sup>	66/154 (43)	48/150 (32)	76/157 (48)	69/153 (45)
3	40 to 45				
	1 <sup>st</sup> lactation	18/90 (20)	34/89 (38*)		
	≥ 2 <sup>nd</sup> lactation	44/160 (28)	38/166 (22)		
4	40-45			154/415 (37)	178/414 (43*)
5	27			132/338 (39)	121/335 (36)
6	28	130/321 (40)	159/313 (51*)		
	56	102/321 (32)	130/313 (42*)		
7	33			28/116 (24)	50/155 (32*)
	61			24/116 (21)	45/155 (29*)
Totals	29 to 45	291/816 (36)	335/809 (41*)	390/1,026 (38)	418/1,057 (40)
	56 to 61	120/412 (29)	171/404 (42*)	...	...

\*Differs ( $P < 0.05$ ) from no-CIDR cows.

<sup>1</sup>Greater ( $P < 0.05$ ) conception rates for Presynch cows. No effect of CIDR insert.

**Table 3. Pregnancy Rates at Day 56-61 After Timed Insemination According to Luteal Activity before Treatment and Concentrations of Progesterone at the Time of PGF<sub>2α</sub> Injection**

Pretreatment cycling status <sup>1</sup>	Study	Serum progesterone before PGF <sub>2α</sub> <sup>2</sup>	No CIDR		CIDR		CL present <sup>3</sup>		
			No CIDR		CIDR				
			-----		No./no. (%) -----				
Noncycling	1	High	19/58 (33)		17/47 (36)		...		
	2	High	8/34 (24)		16/56 (29)		7/25 (28)		
			Subtotal high	27/92 (29)		33/103 (32)			
	1	Low	4/38 (11)		15/41 (37)		...		
	2	Low	2/17 (12)		1/30 (3)		2/12 (17)		
			Subtotal low	6/55 (11)		16/71 (23†)			
Noncycling total			33/147 (22)		49/174 (28)				
Cycling	1	High	69/178 (39)		79/179 (44)		...		
	2	High	14/54 (26)		24/59 (41)		236/651 (36)		
			Subtotal high	83/232 (36)		103/238 (43†)			
	1	Low	10/47 (21)		19/46 (41)		...		
	2	Low	0/11 (0)		4/10 (40)		28/108 (26)		
			Subtotal low	10/58 (17)		23/56 (41*)			
Cycling total			93/290 (32)		126/294 (43*)				

\*Differs ( $P < 0.05$ ) from No CIDR cows.

†Differs ( $P < 0.10$ ) from No CIDR cows.

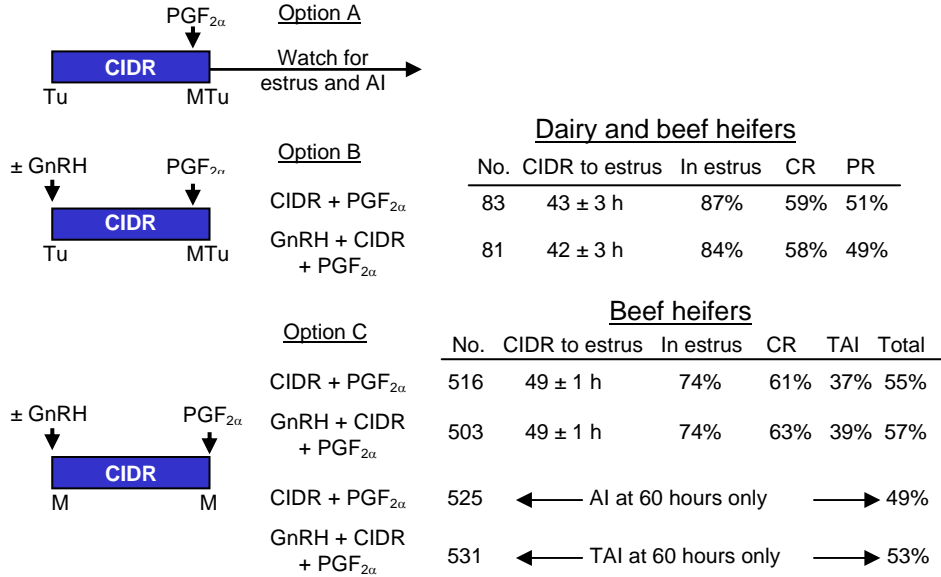
<sup>1</sup> Based on progesterone concentrations measured in 3 blood serum samples collected before each Presynch PGF<sub>2α</sub> injection and before the first GnRH injection of the Ovsynch protocol.

<sup>2</sup> Low = concentrations of progesterone  $< 1$  ng/mL, and high =  $\geq 1$  ng/mL.

<sup>3</sup> Presence of a corpus luteum assessed by transrectal ultrasonography at the first GnRH injection of the Ovsynch protocol. Some CIDR-treated cows may have had elevated progesterone because of the CIDR insert.

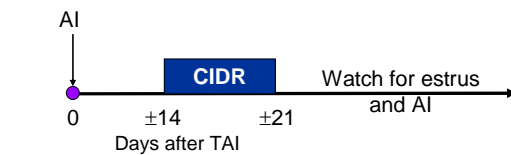


## Replacement Heifer AI-Breeding Options

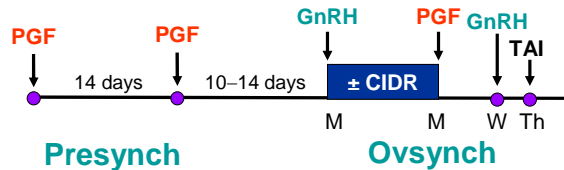


**Figure 1. Various AI Breeding Protocols for Heifers.** Option A is the labeled indication in which PGF<sub>2α</sub> is administered 24 hours before insert removal, and all inseminations are made after detected estrus. Option B tested at 3 locations included or did not include an upfront GnRH injection (CIDR insert to estrus in hours; CR = conception rate; PR = pregnancy rate; Total = combined pregnancy rate for CR + TAI). Option C tested at 12 locations includes insemination after detected estrus (CR) plus a cleanup TAI at 84 hours after insert removal. The last two protocols in Option C are TAI options with no heat detection. AI = artificial insemination; TAI = timed AI; M = Monday; Tu = Tuesday; GnRH = gonadotropin releasing hormone; PGF<sub>2α</sub> = prostaglandin F<sub>2α</sub>; and CIDR = progesterone insert.

### Use of CIDR Insert to Resynchronize Return Estrus



### Use of the CIDR Insert before First AI



**Figure 2. Application of the Progesterone (CIDR) Insert in Lactating Dairy Cows to Resynchronize the Return to Estrus in Cows of Unknown Pregnancy Status or as Part of a First-service AI Breeding Protocol.** AI = artificial insemination; PGF = prostaglandin F<sub>2α</sub>; GnRA = gonadotropin releasing hormone; TAI = timed AI; M = Monday; W = Wednesday; Th = Thursday.