

## Early Detection of Non-Pregnant Holstein Cows Subjected to Artificial Insemination by Administration of Intravaginal Progesterone (CIDR)

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**Abstract:** Artificial insemination breeding programs have long been recommended for dairy producers that raise heifers for herd replacements because of the proven genetic and economic advantages of using AI compared with using natural service bulls for breeding dairy cattle. The aim of this study were to determine the effects of a CIDR insert incorporated into a presynchronized timed AI protocol on detection of estrus, ovulation rate, pregnancy rates and late embryonic loss in high-producing dairy cows. In this study, 60 Holstein dairy cows were allocated into 2 groups of 30 cows. Group 1 (control group) no received any treatments. Group 2 as treatment group received CIDR on day 14 of post AI. Data showed that there is a significant difference among groups in emergence of estrus.

**Key words:** Pregnancy, estrus, cow, progesterone, CIDR, Iran

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### INTRODUCTION

Artificial insemination breeding programs have long been recommended for dairy producers that raise heifers for herd replacements because of the proven genetic and economic advantages of using AI compared with using natural service bulls for breeding dairy cattle. Unfortunately, only 62-68% of dairy heifers on US farms receive at least one AI service (Hogeland and Wadsworth, 1995). When asked to rank reasons for using natural service bulls to breed heifers, farmers listed heifers not at a convenient location, inadequate heat detection for AI and lack of time to supervise AI among the most important factors contributing to this management practice (Erven and Arbaugh, 1987).

Thus, estrus-synchronization protocols that include Timed AI (TAI) to minimize reliance on estrus detection may increase use of AI for breeding dairy heifers (Peeler *et al.*, 2004). Recently, an intravaginal insert impregnated with 1.38 g of progesterone (CIDR, Controlled Internal Drug-releasing Device) was approved for use in lactating dairy cattle in the United States. Use of a CIDR between 14 and 21 days after AI improved the return to estrus of nonpregnant cows (Chenault *et al.*, 2003). Furthermore, incorporation of the CIDR into a timed AI protocol improved pregnancy rates in anovulatory dairy (El-Zarkouny *et al.*, 2004) and beef cows (Lamb *et al.*, 2001). Anovulation by 65 Days in Milk (DIM) affects >20% of the lactating dairy cows in the

United States (Moreira *et al.*, 2001; Cerri *et al.*, 2004; Santos *et al.*, 2004a) with some herds having >40% of the primiparous cows not cycling by 65 DIM (Cerri *et al.*, 2004), leading to reduced conception rates (Rhodes *et al.*, 2003; Cerri *et al.*, 2004; Santos *et al.*, 2004a) and increased embryonic losses (Santos *et al.*, 2004b). Treatment of anovulatory cows with CIDR for 7 days improved detection of estrus, ovulation rate and pregnancy rates (Rhodes *et al.*, 2003) and incorporation of a CIDR into a timed AI protocol using GnRH to induce ovulation improved pregnancy rates.

Dairy cows subjected to a timed AI protocol using Estradiol Cypionate (ECP) to induce ovulation had greater conception and pregnancy rates when observed in estrus at AI which was associated with cyclicity (Pancarci *et al.*, 2002; Cerri *et al.*, 2004). The reseachers hypothesized that incorporating a CIDR to a timed AI protocol using ECP to synchronize ovulation would benefit cows by improving detection of estrus and ovulation resulting in greater pregnancy rates. Therefore, the objectives of the current study were to determine the effects of a CIDR insert incorporated into a presynchronized timed AI protocol on detection of estrus ovulation rate, pregnancy rates and late embryonic loss in high-producing dairy cows.

### MATERIALS AND METHODS

In this study, 60 Holstein dairy cows were allocated into 2 groups of 30 cows. Group 1 (control group) no

received any treatments. Group 2 as treatment group received CIDR on day 14 of post AI. While 7 days after to wit on days 21 of post AI, all CIDRs were excised and on day 30 were assayed to pregnancy possibility. This assessment was done by an ultrasonography modeled Honda-HS 1500 with 7.5 MHz probe.

**RESULTS AND DISCUSSION**

Results showed that in group 2, 14 of understudying cows were pregnant which not showed estrus signs after excision of CIDR and 10 of them were non-pregnant which have showed estrus signs after excision of CIDR. In group 1, 12 of them were pregnant which did not show estrus and 6 of them which have sowed estrus signs were non-pregnant. In the treatment group, 1 of cattle, despite showing signs of estrus was pregnant and 2 of them despite not showing signs of estrus were not pregnant. In the control group, 12 of livestock's, despite not showing signs of estrus were not pregnant (Table 1-4). It must be remember that in treatment group, 3 of animals due to prematurely exit the CIDR were excluded.

This study revealed that use of progesterone as CIDR on day 14 and excision of it on day 21 yields to early detection of pregnancy. As respects that most of fetal deaths occurs on day 14 after insemination, thus most of them not show any pregnancy signs until day 31 by ultrasonography. In one study, gradual increase in progesterone and decrease in total progesterone in cows which have fertility problems has been proved on day 6 after estrus and evidence showed that the most of fetal deaths in cattle is due to inadequate secretion of the

corpus luteum in early pregnancy (Bage *et al.*, 2002). The idea which use of progesterone on days 9-5 after insemination was to prevent a possible diminish of progesterone during the 1st week after insemination (Helmer and Britt, 1986), improvement in embryo development (Garrett, 1988), secretion of interferon (Kerbler *et al.*, 1997) and to stimulate uterine to secretions necessary for embryo development (Nation *et al.*, 2000).

Anovulation in the 1st 65 days postpartum affects >20% of the lactating dairy cows in the United States (Moreira *et al.*, 2001; Cerri *et al.*, 2004; Santos *et al.*, 2004a) with some herds having >40% of the primiparous cows not cycling by 65 DIM (Cerri *et al.*, 2004), leading to reduced conception rates (Rhodes *et al.*, 2003; Cerri *et al.*, 2004; Santos *et al.*, 2004a) and increased embryonic losses (Santos *et al.*, 2004b). Treatment of anovulatory cows with progesterone before 1st postpartum ovulation minimized the occurrence of short luteal cycles and improved conception rates (Inskeep, 2002). The loss of 2.6% of the CIDR inserted is similar to the 2.7% reported by Chenault *et al.* (2003).

As expected, the CIDR insert eliminated estrus behavior before its removal and tended to decrease ovulation in the 1st 48 h after the last PGF2 $\alpha$  treatment because of the negative feedback of progesterone on LH secretion which prevents estrus and the LH surge. The smaller diameter of the dominant follicle for the CIDR group at the last PGF2 $\alpha$  injection was observed mainly because of a decrease in follicle size in primiparous cows. This effect may be attributed to an inhibitory effect of progesterone on LH pulse frequency when CIDR is administered (Burke *et al.*, 1996) which might have been more pronounced in primiparous cows due to the lower milk yield and consequent reduced clearance of progesterone (Sangsrivavong *et al.*, 2002). This could explain the effect of parity on follicle diameter at the final injection of PGF2 $\alpha$  and 48 h later, however when the CIDR was removed, follicle growth and diameter before ovulation were similar for both treatments indicating that follicle growth was resumed similarly between groups when the CIDR was no longer inserted. A 476 cows with high progesterone levels at the GnRH, only 6.7% had premature spontaneous CL regression during the timed AI protocol.

This low proportion was expected because cows had their estrous cycles presynchronized with two injections of PGF2 $\alpha$  (Moreira *et al.*, 2001), however more CIDR-treated than control cows experienced premature spontaneous CL regression. It is not clear why CIDR-treated cows had increased spontaneous CL regression during the timed AI protocol but it is possible that the presynchronization with PGF2 $\alpha$  was not as effective in

Table 1: Evaluation of return to estrus in the treatment group

Treatment groups	No.	Percentage
Return to estrus	11	40.7
Not return to estrus	16	59.3
Total	27	100.0

Table 2: Evaluation of pregnancy in the treatment group

Treatment groups	No.	Percentage
Pregnant	15	44.4
Non-pregnant	12	55.6
Total	27	100.0

Table 3: Evaluation of return to estrus in the control group

Treatment groups	No.	Percentage
Return to estrus	6	20
Not return to estrus	24	80
Total	30	100

Table 4: Evaluation of pregnancy in the control group

Treatment groups	No.	Percentage
Pregnant	12	40
Non-pregnant	18	60
Total	30	100

cows in the CIDR compared to those in the control group. Although, ovulatory response to GnRH was not evaluated in all cows those with progesterone  $<1 \text{ ng mL}^{-1}$  at the GnRH injection and treated with a CIDR insert had a decreased incidence of CL at the final injection of PGF2 $\alpha$  compared with control cows with progesterone  $<1 \text{ ng mL}^{-1}$  at the GnRH. The decreased ovulation to GnRH in low-progesterone cows when treated with CIDR might be related to the negative feedback of progesterone on LH secretion.

When ovariectomized cows received a CIDR insert, LH concentrations and pulse frequency were decreased in the 1st 8 h of insert administration (Burke *et al.*, 1996). However, ovulation to GnRH, based on changes in progesterone from the day of GnRH treatment and 7 days later did not differ in dairy (El-Zarkouny *et al.*, 2004) or beef cows (Lamb *et al.*, 2001) when treated with a CIDR. The decreased ovulation rate to the GnRH and the greater premature luteolysis resulted in a reduced proportion of cows in the CIDR-treated group with a CL at the PGF2 $\alpha$  treatment of the timed AI protocol.

The presence of CL at PGF2 $\alpha$  treatment of timed AI protocols has been shown to positively affect pregnancy rates (Moreira *et al.*, 2001; Cerri *et al.*, 2004) which was also observed in the current study. Incorporation of a CIDR insert into a timed AI protocol using ECP to induce ovulation did not influence reproductive variables in lactating dairy cows. Control and CIDR-treated cows had similar detection of estrus, pregnancy rates and late embryonic losses. When the CIDR was incorporated into the ovsynch (day GnRH, PGF2 $\alpha$  and 9 GnRH and timed AI 12-20 h after GnRH) protocol, pregnancy rates were improved on day 28 of gestation for anovulatory cows and on day 57 for all cows (El-Zarkouny *et al.*, 2004). However when cows were presynchronized with PGF2 $\alpha$  incorporation of a CIDR insert into the ovsynch, protocol did not benefit pregnancy rates and embryo survival (El-Zarkouny *et al.*, 2004). It is possible that the benefits from incorporation of a CIDR insert into ovsynch timed AI protocol were related to preventing cows from prematurely coming into estrus and ovulating, thereby improving synchrony of ovulation and conception at timed AI.

Another explanation for lack of response to the CIDR in the current study was the fact that cows were inseminated the day before scheduled timed AI if observed in estrus.

When incorporation of a CIDR insert demonstrated benefits in fertility, cows were inseminated at fixed-time, 12-20 h after the final GnRH of the ovsynch which requires optimal synchronization after the GnRH-induced ovulation because of lack of estrus detection.

## CONCLUSION

Incorporation of an intravaginal progesterone insert into a presynchronized timed insemination protocol using ECP to induce estrus and ovulation did not improve detection of estrus, ovulation rate, pregnancy rates and late embryonic loss in high-producing lactating dairy cows. Cows detected in estrus had improved pregnancy rates and decreased embryonic losses due to greater ovulation rate which was influenced by ovulatory follicle size. Anovulatory cows subjected to a presynchronized timed AI protocol using ECP had similar ovulation rates but decreased detection of estrus and pregnancy rates and greater late embryonic loss than cyclic cows. Therefore incorporation of a progesterone insert to a presynchronized timed AI protocol using ECP did not improve reproductive performance of lactating dairy cows regardless of cyclic status. Improvements in pregnancy rates are expected when display of estrus is increased in the 1st 48 h after treatment with ECP.

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