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Pregnancy Rates in Lactating Dairy Cows After Presynchronization of Estrous Cycles and Variations of the Ovsynch Protocol*

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ABSTRACT

Our objectives were to determine pregnancy rates after altering times of the second GnRH injection, insemination, or both in a combined Presynch + Ovsynch protocol, to accommodate once-daily lockup of dairy cows. Lactating dairy cows (n = 665) from 2 dairy herds in northeastern Kansas were studied. Cows ranged from 24 to 44 d in milk (DIM) at the start of the Presynch protocol, which consisted of 2 injections of $PGF_{2\alpha}$ 14 d apart, with the second injection given 12 d before initiating the Ovsynch protocol. Cows were blocked by lactation number and assigned randomly to 3 treatments consisting of variations of the Ovsynch protocol. Cows in 2 treatments received injections of GnRH 7 d before and 48 h (G48) after the $PGF_{2\alpha}$ injection. Timed AI (TAI) was conducted at the time of the second GnRH injection (G48 + TAI48) or 24 h later (G48 + TAI72). Cows in the third treatment received the injections of GnRH 7 d before and at 72 h after $PGF_{2\alpha}$ and were inseminated at the time of the second GnRH injection (G72 + TAI72). Pregnancy was diagnosed weekly by palpation per rectum of uterine contents on d 40 or 41 after TAI. Pregnancy rates differed between herds, but they were consistently greater for G72 + TAI72 than for G48 + TAI48 and G72 + TAI72. Subsequent calving rates were consistent with differences in initial TAI pregnancy rates. Pregnancy loss was least for cows on the G72 + TAI72 treatment. Body condition scores (BCS) ranged from 1.0 to 4.0 when assessed on Monday of the breeding week. An interaction of BCS and herd was detected in which cows in herd 1 having poorer BCS (<2.25) had greater pregnancy rates than cows of greater BCS (≥2.25), whereas the reverse was true in herd 2 in which overall pregnancy rates were greater. We concluded that inseminating at 48 or 72 h after $PGF_{2\alpha}$, when GnRH was administered at 48 h after $PGF_{2\alpha}$, produced fewer pregnancies than inseminating and injecting GnRH at 72 h after $PGF_{2\alpha}$ for cows whose estrous cycles were synchronized before initiating this variant of the Ovsynch protocol.
(**Key words:** Ovsynch, Presynch, pregnancy rate)

Abbreviation key: CL = corpus luteum, **G48 + TAI48** = GnRH injection and TAI at 48 h after $PGF_{2\alpha}$, **G48 + TAI72** = GnRH injection at 48 h and TAI at 72 h after $PGF_{2\alpha}$, **G72 + TAI72** = GnRH injection and TAI at 72 h after $PGF_{2\alpha}$, **Ovsynch** = injection of GnRH 7 d before and 48 h after $PGF_{2\alpha}$ before TAI, **P4** = progesterone, **Presynch** = 2 injections of $PGF_{2\alpha}$ 14 d apart with the second injection given 12 d before initiating the Ovsynch protocol, **TAI** = timed AI.

INTRODUCTION

Estrus-detection efficiency (AI submission rate) is one of the most important factors limiting reproductive performance of dairy cows. Advances in genetics and management have changed the dairy industry in the face of increasing numbers of cows per herd (Lucy, 2001). The shift in herd size has created a new problem for reproductive management of dairy cows. Traditional methods of detecting estrus are inefficiently applied in large dairy herds in which the number of cows managed per worker has increased, resulting in decreased accuracy and efficiency of detecting estrus. According to recent studies, <50% of the lactating dairy cows were detected in estrus (Senger, 1994; Washburn et al., 2002), resulting in prolonged interinsemination intervals (Stevenson et al., 1983) and less profit for the dairy producer.

One way to manage the decline in reproductive performance of dairy herds is to apply methods to control follicular dynamics and luteal life span. Application of the **Ovsynch** protocol [injection of GnRH 7 d before and 48 h after $PGF_{2\alpha}$ before timed AI (**TAI**)] allows for synchronization of follicular development, luteal regression, and time of ovulation with minimal need for detection of estrus (Pursley et al., 1995, 1997a,b). Conception rates after TAI in some studies are less than those when AI is based on detected estrus (Burke et al., 1996; Pursley et al., 1997a,b; Stevenson et al., 1999; Jobst et al., 2000), but because more cows are inseminated, depending on estrus detection or AI sub-

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mission rates, pregnancy rates are usually greater after TAI (Pursley et al., 1997a,b; Cartmill et al., 2001b).

Stage of the estrous cycle at which the Ovsynch protocol is initiated influenced its effectiveness. Initiating the protocol in cows during early to middiestrus (d 5 and 12) produced greater pregnancy rates than those in which the Ovsynch protocol was initiated on other days of the estrous cycle (Vasconcelos et al., 1999). One way to target the initiation of this protocol during the most favorable stage of the estrous cycle is to presynchronize the estrous cycles of cows before the first injection of GnRH of the Ovsynch protocol (Moreira et al., 2001; El-Zarkouny et al., 2004). Use of 2 injections of $PGF_{2\alpha}$, administered 14 d apart, with 12 intervening d before initiation of Ovsynch (**Presynch**), constitutes a practical and relatively inexpensive method for presynchronization. More than 70% of the cows began the Ovsynch protocol during early- to middiestrus (between d 5 and 12) when Presynch was applied before Ovsynch, compared with 53% of the cows treated with Ovsynch at random stages of the cycle (El-Zarkouny et al., 2004).

Establishing the optimal time for TAI after the second GnRH injection of the Ovsynch protocol was part of the original Ovsynch study. Lactating dairy cows were inseminated at the same time as GnRH was administered (48 h after $PGF_{2\alpha}$) or at 8, 16, 24, or 32 h after GnRH (Pursley et al., 1998). Although pregnancy rates were similar for cows inseminated from 0 to 24 h, and cows inseminated at 32 h after GnRH had reduced pregnancy rates, pregnancy outcomes at 0 to 32 h produced a quadratic response curve, indicating that insemination at 16 h was optimal. Inseminating at 16 h after GnRH, however, is inconvenient because it may not coincide with once-daily lockup of cows at the feed bunk after the morning milking. Most importantly, timing of AI for cows pretreated with the Presynch protocol before Ovsynch has not been addressed.

When we consider the increased proportion of cows between d 5 to 12 of the estrous cycle after a combined Presynch + Ovsynch protocol and more uniformity of follicular dynamics in synchrony with luteal demise after $PGF_{2\alpha}$, timing of the second GnRH injection and TAI may be different from inseminations made between 0 and 24 h after GnRH in the Ovsynch protocol. Therefore, the objective of the present experiment was to determine pregnancy rates after altering times of the second GnRH injection, insemination, or both in a combined Presynch + Ovsynch protocol, to correspond with once-daily lockup of dairy cows.

MATERIALS AND METHODS

Herd Management

Lactating dairy cows (n = 715) calving between July 2002 and May 2003 were used at 2 cooperating commercial dairy herds in northeastern Kansas. The herds were composed of 628 to 696 cows and were milked 3 times daily, with rolling herd averages ranging from 11,800 to 12,400 kg of milk. Cows were housed in either 2- or 4-row free-stall barns consisting of feed-line head lockups and free stalls bedded with sand, equipped with sprinklers and fans above the feed line and fans above the free stalls. Cows were fed a TMR at least twice daily, consisting of corn silage, chopped alfalfa, whole cottonseed, corn or milo grain, soybean meal, vitamins, and minerals. Cows had free access to fresh water ad libitum in open tanks at 3 or more locations in each 100-cow pen.

Experimental Design

Cows were grouped into breeding clusters every 2 wk within each herd. The DIM ranged from 24 to 44 d at the start of the Presynch protocol. All cows received two 25-mg injections of $PGF_{2\alpha}$ (5 mL of Lutalyse; Pharmacia Animal Health, Kalamazoo, MI) 14 d apart, with a second injection given 12 d before initiating the Ovsynch protocol. Cows were blocked by lactation number (1 vs. 2+) and assigned randomly to 3 treatments, consisting of variations in the timing of GnRH and AI after the $PGF_{2\alpha}$ injection of the Ovsynch protocol. Inseminations were performed between September 2002 and June 2003. Insemination was carried out between 65 and 79 DIM in one herd and between 58 and 79 DIM in the second herd. Treatments were initiated every 2 wk within herd, with cows from each herd being inseminated on alternate weeks. Body condition score (1 = thin to 5 = fat) was evaluated on Monday of the breeding week.

Treatment protocols are illustrated in Figure 1. Cows in 2 treatments received one 100-μg injection of GnRH (2 mL of Fertagyl; Intervet, Millsboro, DE) 7 d before and 48 h after a $PGF_{2\alpha}$ injection. Fixed TAI occurred concurrent with the second GnRH injection (**G48 + TAI48**) or 24 h later (**G48 + TAI72**). Cows in the third treatment received the first injection of GnRH 7 d before, and the second injection of GnRH at 72 h after $PGF_{2\alpha}$, concurrent with TAI (**G72 + TAI72**). All cows in the experiment were treated and inseminated during the morning hours (0700 to 1000 h) after milking, while restrained in feed-line head lockups. Cows that were detected in estrus before their scheduled TAI were not inseminated until their scheduled breeding time. Pregnancy was diagnosed weekly by palpation per rectum of uterine contents on d 40 (herd 1) or 41 (herd 2) by the same veterinary practitioner.

In one herd (herd 2) during November to January, representing 5 breeding clusters of 117 cows, observations of sexual behavior were recorded based on daily-