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Pregnancy outcomes are not improved by administering gonadotropin-releasing hormone at initiation of a 5-day CIDR-Cosynch resynchronization protocol for lactating dairy cows

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ABSTRACT

Using a 5-d controlled internal drug-release (CIDR)-Cosynch resynchronization protocol, the objective of this study was to determine the effect of the initial GnRH injection on pregnancy per artificial insemination (P/AI) to the second artificial insemination in lactating Holstein dairy cows. On 37 ± 3 d (mean ± standard deviation) after the first artificial insemination, and upon nonpregnancy diagnosis (d 0 of the experiment), lactating cows eligible for a second artificial insemination (n = 429) were enrolled in a 5-d CIDR-Cosynch protocol. On d 0, all cows received a CIDR insert and were assigned randomly to receive the initial GnRH injection (GnRH; n = 226) of the protocol or no-GnRH (n = 203). Blood samples were collected from a sub-group of cows (n = 184) on d 0 and analyzed for progesterone (P4) concentration. On d 5, CIDR inserts were removed, and all cows received 1 injection of $PGF_{2\alpha}$. On d 6 and 7, cows were observed once daily by employees for tail-chalk removal, and cows detected in estrus on d 6 or 7 received artificial insemination that day (EDAI), and did not receive the final GnRH injection. The remaining cows not detected in estrus by d 8 received GnRH and timed artificial insemination (TAI). Pregnancy status was confirmed by transrectal palpation of uterine contents at 37 ± 3 d (mean ± standard deviation) after the second artificial insemination. Eliminating the initial GnRH injection had no effect on P/AI compared with cows receiving GnRH (27 vs. 21%), respectively. Similarly, method of insemination (EDAI vs. TAI) and its interaction with treatment had no effect on P/AI. Primiparous cows had greater P/AI than multiparous cows (31 vs. 21%). Mean P4 concentrations (n = 184) at the initiation of the protocol did not differ between treatments (4.51 ± 0.35 ng/mL no-GnRH vs. 3.96 ± 0.34 ng/mL of GnRH). When P4 concentrations were categorized as high (≥1 ng/mL) or low (<1 ng/mL), P/AI tended to be greater for high P4 concentrations (n = 136) compared with low (n = 48) P4 concentrations (26 vs. 16%, respectively). No differences were observed in the proportion of cows with high or low P4 between treatments. Collectively, these results provide evidence that eliminating the initial GnRH in a 5-d CIDR-Cosynch resynchronization protocol for lactating dairy cows did not reduce P/AI in this study.

Key words: gonadotropin-releasing hormone, resynchronization, dairy cow

INTRODUCTION

It is well documented that approximately 60% of dairy cows fail to conceive to the first AI and will require a second or more inseminations (Fricke et al., 2003; Galvão et al., 2007; Bisinotto et al., 2010). Given that detection of estrus is a challenge on many dairies, and pregnancy per AI (**P/AI**) to second and greater timed-AI (**TAI**) is less than first AI (Fricke et al., 2003; Chebel et al., 2006; Sterry et al., 2007), the need to develop or modify resynchronization programs in lactating dairy cows is warranted.

Pregnancy to a TAI protocol is greater if cows respond to the initial GnRH with ovulation of a dominant follicle, complete luteolysis by the time of AI, and exhibit synchrony of ovulation at the end of the protocol (Santos et al., 2010; Bisinotto et al., 2014; Stevenson, 2016). Occurrence of anestrus and anovulation, asynchrony of return to estrus after first AI, and even embryonic loss, however, create a milieu of ovarian structures and hormonal profiles that may reduce the desired physiological responses to exogenous hormones in resynchronization protocols (Lopes et al., 2013; Wijma et al., 2016; Ricci et al., 2017). For example, when GnRH-based synchronization protocols are initiated during random stages of the estrous cycle, 10 to 40% of dairy cows fail to synchronize with ovulation because of

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variation in ovarian structures at the time of protocol initiation (Pursley et al., 1995; Vasconcelos et al., 1999; Stevenson, 2016).

Failure of ovulation to the first GnRH injection can reduce the success of TAI protocols and attainment of pregnancy (Vasconcelos et al., 1999; Carvalho et al., 2015; Stevenson, 2016). It is plausible that the variation of ovarian structures and hormonal profiles among cows failing to conceive to a previous AI, and subjected to resynchronization, may result in ovulation failure after the initial GnRH injection, and could contribute to the suboptimal results achieved with current resynchronization methods without presynchronization. In fact, Bisinotto et al. (2010) demonstrated only 35% of dairy cows ovulated in response to the initial GnRH when subjected to an Ovsynch or 5-d controlled-internal drug release (**CIDR**)-Ovsynch resynchronization protocol.

The requirement of the initial GnRH injection has been examined in CIDR-based TAI protocols. Currently, reported results are inconsistent when beef and dairy cattle are subjected to a 5-d CIDR-Cosynch TAI protocol with or without the initial GnRH. The results of several studies provide evidence that inclusion of the initial GnRH injection in dairy heifers (Colazo and Ambrose, 2011; Lima et al., 2011; Howard et al., 2012) and beef heifers (Howard et al., 2009; Cruppe et al., 2014) does not improve P/AI. In contrast, Lima et al. (2013) and Kasimanickam et al. (2014) reported an improvement in P/AI when the initial GnRH was included in a 5-d CIDR-Cosynch for dairy and beef heifers. The focus of the previous 2 studies, however, was to determine the effect of 1 versus 2 $PGF_{2\alpha}$ injections, and not the role of the initial GnRH injection.

Studies in beef cows (Lamb et al., 2006) and dairy cows (Colazo and Ambrose, 2015) have shown no advantage in P/AI following administration of GnRH at CIDR insertion. To our knowledge only one study has examined the effect of the initial GnRH in a 5-d CIDR TAI protocol in lactating dairy cows. Colazo and Ambrose (2015) observed no difference in P/AI with or without the initial GnRH in a 5-d CIDR-Ovsynch resynchronization protocol in lactating dairy cows. Nevertheless, the previously mentioned study (Colazo and Ambrose, 2015) did not specifically investigate the necessity of the initial GnRH; rather, the focus was on the number of $PGF_{2\alpha}$ injections following CIDR removal in lactating dairy cows.

It remains unclear whether use of GnRH at the time of CIDR insertion in a 5-d CIDR-Cosynch protocol is necessary in lactating dairy cows. More importantly, use of GnRH and its effectiveness at the initiation of this type of resynchronization protocol in dairy cows has not been exclusively examined. Therefore, we hypothesized that similar P/AI would be achieved with or without the initial GnRH injection for dairy cows subjected to a 5-d CIDR-Cosynch resynchronization protocol with one injection of $PGF_{2\alpha}$ following CIDR removal. The specific objective of this study was to determine the effect of the initial GnRH injection in a 5-d CIDR-Cosynch resynchronization protocol on P/AI for the second insemination in lactating Holstein dairy cows.

MATERIALS AND METHODS

All animal handling procedures and treatment protocols were approved by the Animal Care and Use Committee at the University of Idaho. This study was conducted on a commercial dairy farm in Pasco, Washington, from January through July 2014. The dairy farm milked 5,000 Holstein cows, with an average 150-d ECM production of 35 kg/cow per d. All cows were housed in freestall barns, milked twice daily, and a TMR was fed twice daily (morning and evening) with feed pushups every 6 h. Cows also had ad libitum access to water and >20 h access to TMR. The TMR was formulated to meet or exceed the nutritional requirements for high-producing cows (NRC, 2001), and cows were not supplemented with recombinant bST. The voluntary waiting period was 50 d, and estrous cycles were synchronized for first postpartum AI using a Presynch-Cosynch protocol ($PGF_{2\alpha}$ – 14 d – $PGF_{2\alpha}$ – 11 d – GnRH – 7 d – $PGF_{2\alpha}$ – 72 h – GnRH and AI). Cows detected in estrus after the second $PGF_{2\alpha}$ injection of the Presynch protocol, or after the $PGF_{2\alpha}$ injection of the Cosynch protocol, were inseminated and did not complete the Cosynch protocol. The average DIM to first breeding (**DIMFB**) was 64 d. The average P/AI for first insemination was 26%.

Experimental Protocol and Treatment

Between 35 to 42 d (37 ± 3 d, mean ± SD) after the first AI, cows were subjected to weekly pregnancy diagnosis by the herd veterinarian by transrectal palpation of uterine contents. After nonpregnant diagnosis for first service, 476 Holstein cows were initially enrolled into this experiment. Forty-seven cows, however, were eliminated from this study for reasons such as sold/died before pregnancy diagnosis after treatment, or because of a discrepancy in farm records. Therefore, 429 cows completed the study and were used for the analysis on the effect of the initial GnRH in a 5-d CIDR-Cosynch resynchronization on P/AI for second insemination.

On d 0 of the experiment (the day of nonpregnancy diagnosis after first insemination), cows were assigned randomly to receive the initial GnRH (GnRH) injection (n = 223, i.m., 100 μg of gonadorelin hydrochloride;