Reproductive parameters in Holstein dairy cows treated with three resynchronization methods

Nasroallah Moradi kor 1*, Kaveh Mohammadi Khanghah 2, Mona Ghasemi 2

1 Department of Animal Science, Islamic Azad University, Baft Branch, Baft, Iran, 2 Department of Animal Science, Razi University of Agricultural Sciences and Natural Resources, Kermanshah, Iran

Abstract: The objective of the present study was to determine the reproductive parameters in Holstein cows treated by different hormonal protocols during insemination and postpartum periods. The 114 dairy cows (multiparous) with BCS between 2.75 and 3 were divided into 3 groups in a completely randomized design. 38 cows were treated by Heatsynch method + eCG (M1), 38 cows received Ovsynch + CIDR (M2) and 38 cows were subjected to the Heatsynch method (M3). Blood samples were taken to determine progesterone plasma contents in 14, 21 and 24 days after first insemination. Results showed that treatment with M3 protocol significantly decreased days to first service and conception rate in the cows (P < 0.05). First service conception rate in cows treated with M1 protocol was higher (P < 0.05) compared to M2 and M3 protocol. Pregnancy rate in M1 was significantly higher than that of the other groups (P ≤ 0.05). This difference could be due to equine chorionic gonadotropin (eCG) which could have an effective function on ovulation, fertilization and embryo's vitality. Plasma progesterone levels of pregnant cows were higher than that of nonpregnant cows regardless the type of resynchronization protocols (P≤0.05). However, activation of ovaries, reinitiate estrous cycle and accordance between estrous and ovulation were increased when dairy cows subjected to the Heatsynch + eCG method (M1). Thus, M1 protocol is recommended for primiparous but not for multiparous cows, and T3 protocol is not recommended for synchronization.

Key words : Progesterone, Reproductive indices, CIDR, Dairy cattle, eCG

I. Introduction

Lactating dairy cows with high genetic merit and outstanding milk production are likely to be more vulnerable to fertility problems, such as lower AI conception rates, weaker expression of estrus, and greater embryonic loss after insemination than lower producing cows [18]. Reproductive efficiency in dairy herds increases by inseminating all the cows shortly after the end of the voluntary waiting period, obtaining high pregnancy rate to first service, enhancing embryonic and fetal survival, and detecting and re-inseminating non-pregnant cows. Ultrasonography enables early pregnancy diagnosis and detection of non-pregnant cows, which can be subjected to resynchronization of ovulation and timed AI (TAI) to minimize the problem of low estrus detection [24]. When estrus was synchronized and AI was scheduled after detected estrus in earlier studies, fertility was greater than when AI was made at fixed times after synchronization without regard to detected estrus [4, 19, 16]. Poorer fertility after timed AI (TAI) often was attributed to insufficient synchrony of estrus and ovulation to allow appropriate timing of AI relative to ovulation [8]. Deficiencies in luteal function [6], either before or after insemination, are associated with reduced fertility in beef [21] and dairy cattle [10]. In addition, concentrations of progesterone (P4) in blood 34 to 48 h before the preovulatory surge of LH were greater in cows that conceived compared with those that failed to conceive [7]. Thus, the magnitude of P4 concentrations before estrus may be associated with factors that increase the probability of conception. Blood concentrations of P4 during the luteal phase before insemination are associated positively with conception rate [9, 7, 12, 10, 26, 27]. Increased pregnancy rate was reported after intra vaginal inserts containing P4 were applied to cows that were synchronized with PGF2α [33]. Therefore, reproductive performance of cows receiving the Ovsynch protocol may be improved if P4 is administered during the 7 d between the first GnRH and the only PGF2α injections. Progesterone should prevent premature estrus and ovulation during the period in which spontaneous luteolysis may occur in small percentages of cows whose dominant follicles are not responsive to the first GnRH injection [30, 25, 28, 31, 32]. Protocols for synchronization of ovulation and TAI, such as Ovsynch (GnRH on day 0, PGF 2α on day 7, GnRH on day 9 and TAI 16 h later; [25, 1997] and Heatsynch (GnRH on day 0, PGF 2α on day 7, ECP on day 8, and TAI on day 10; [22], insure that all cows are inseminated and generate acceptable pregnancy rates in lactating dairy cows. Since the stage of the estrous cycle is known to affect the response to these protocols [31, 20], a presynch program was developed that allows initiation of Ovsynch in the early- to mid-luteal phase of the estrous cycle, improving pregnancy rate to first service in cyclic cows [20]. Protocols for resynchronization of estrus that considered the stage of the estrous cycle have been applied after detection of non-pregnant cows by per rectum palpation of
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the uterus and assignment of protocols based on the presence or absence of a CL [1]. Another approach for resynchronization after non-pregnancy diagnosis by ultrasonography was either to initiate the Ovsynch protocol 7 days before ultrasonography or to take advantage of a natural resynchronization after previous service with application of shortened protocols that used PGF$_{2\alpha}$ to induce luteolysis and then GnRH [29] or ECP [3] to induce ovulation. An alternative is to consider the stage of the estrous cycle at the time of initiation of the resynchronization protocol by evaluating ovarian structures and uterine characteristics at the time of a non-pregnancy diagnosis. Determination of the stage of the estrous cycle can be done using per rectum examination and ultrasonography [24] of the genital tract. The objective of this study was to effects of different resynchronization methods on reproductive indices in lactating dairy cows.

II. Materials and methods

This study was performed at a private dairy farm housing consisting 300 Holstein Friesian milking cows with cooperation Rezvan Agriculture junior college located in Kerman province (latitude 25° 55’ N, longitude 53° 26’ E, altitude 1755m) in Iran. Cows were housed in free stall barns and dry lots and fed a total mixed ration (TMR) thrice daily to meet or exceed requirements for lactating cows. Cows with reproductive abnormalities (i.e., metritis, pyometra, uterine or ovarian adhesions) were not included in the study. The 114 dairy cows (second lactation) with BCS between 2.75 and 3 were divided into 3 groups in a completely randomized design. 38 cows were treated by Heatsynch method + eCG (T1), 38 cows received Ovsynch + CIDR (T2) and 38 cows were subjected to the Heatsynch method (T3). Group M1 (n= 38): Cows in the Heatsynch+ eCG group received 100 mg im of GnRH and 200 (IU) eCG on day 0, 25 mg im of PGF$_{2\alpha}$ on day 7, 1 mg im of ECP(0.5 mL of ECP1 sterile solution; Pfizer Animal Health) on day 8, and TAI 48 h later (day10).

Fig. 1. Heatsynch + eCG protocol.

Group M2 (n= 38): Cows in the Ovsynch + CIDR group received 100 mg im of GnRH (2 mL of Cystorelin1; Merial Ltd., Iselin, NJ, USA) on day 0, 25 mg im of PGF$_{2\alpha}$ (5 mL of Lytalyse1 sterile solution; Pfizer Animal Health, New York, USA) on day 7, 100 mg im of GnRH on day 9, and TAI 16 h later (day 10).

Fig. 2. Ovsynch + CIDR protocol.

Group M3 (n= 38): Cows in the Heatsynch group received 100 mg im of GnRH on day 0, 25 mg im of PGF 2a on day 7, 1 mg im of ECP(0.5 mL of ECP1 sterile solution; Pfizer Animal Health) on day 8, and TAI 48 h later (day10).

Fig. 3. Heatsynch protocol.

In days 14, 21 and 24 after insemination, blood samples were taken from jugular vein in 10-ml vacuum tubes (venoject) for pregnancy diagnosis. Serum was recovered by centrifugation (10 minutes at 3000 rpm) and stored at -20°C until assayed for Serum progesterone concentrations using commercially available ELISA kit (Demeditec Diagnostics GmbH, Kiel, Germany). The reproductive parameters recorded. Data was analysed
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using the General Linear Model (GLM) procedures of statistical analysis system (SAS) by completely randomize design and the means were compared using Duncan´s multiple range test.

III. Results and Discussion

The results of reproductive parameters are summarized in (Table 1). The number of S/C for pregnant cows was lower for cows treated with M2 protocol compared to M1 and M3 protocols respectively; \( P < 0.01 \). No difference \( (P > 0.05) \) was observed for Service/Conception (S/C) between the cows with M2, M2 and M3 protocols. First service conception rate on Day 30 for cows in the M1 protocol tended to be higher than those for cows in the M2 and M3 protocols. However, there was a significantly difference \((P< 0.05)\) for FSCR amongst M1 with M2 and M3 protocols.

Table 1. The effect of three resynchronization methods on reproductive parameters of dairy cows.

<table>
<thead>
<tr>
<th>Parameter(s)</th>
<th>Heatsynch + eCG</th>
<th>Ovsynch + CIDR</th>
<th>Heatsynch</th>
</tr>
</thead>
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<tr>
<td>RS</td>
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<td>%35</td>
<td>%30</td>
</tr>
<tr>
<td>OS</td>
<td>%55</td>
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<tr>
<td>%73</td>
<td>%50</td>
<td>%56</td>
<td></td>
</tr>
<tr>
<td>2/S/C</td>
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<td>1/98 ± 0.19</td>
<td>2/33 ± 0.25</td>
</tr>
<tr>
<td>Open days</td>
<td>104</td>
<td>96</td>
<td>121</td>
</tr>
</tbody>
</table>

1First service conception rate, 2Second service conception rate, 3Third service conception rate, 4Service/Conception, 5Return Scale, 6Obtain Scale

Synchronization of oestrus using prostaglandin programs in lactating cows is an established technique for reducing the post partum interval to first insemination [15, 23]. The results from this study showed that treatment with PGF2α on Day 15 post partum has the potential to reduce days to conception and number of S/C only in cows that were treated with M2 protocol (Table 1). These results are consistent with observations reported by Bennrad & Stevenson, [2] and Young et al. [34], who indicated that a single intramuscular injection of PGF2α in the early post partum period reduced the post partum interval to conception. However, our results are in contrast with the results of López-Gatius and Camón-Urgel, [17], who reported that interval from calving to conception, number of services per conception and percentage of cows pregnant to first service were not influenced by treatment of PGF2α on Day 14 post partum. Our results showed that treatment with PGF2α on Day 15 post partum significantly improved FSCR only in cows were treated with M1 protocol (Table 1). Results of this experiment revealed that Plasma progesterone in first group was significantly different than that of the other groups (Fig. 4).

![Fig.4. Mean blood progesterone concentration in days after AI.](image-url)
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cytotoxicity [11]. In the present study, administration of GnRH on Day 23 post-AI increased the days to conception and the number of S/C and decreased FSCR in cows treated with M₁ compared to M₂ protocol (Table 1). Several studies have investigated the effect of treatment with GnRH or equine chorionic gonadotropin (eCG) after insemination to improve fertility by inducing ovulation and forming an accessory corpus luteum [13]. Corpus luteum development results in secretion of P₄, which influences embryo development, interferon-τ production, and inhibition of the luteolytic cascade. Administration of GnRH on days 22–23 after a previous AI should induce ovulation in the majority of cows and result in formation of a CL and synchronization of an ovarian follicular wave. Observations reported by Chebel et al. [3], indicated that the administration of GnRH on Day 21 after AI to lactating dairy cows of unknown pregnancy status, did not affect pre-enrollment pregnancy rates determined on Days 28 and 42 after insemination.

IV. Conclusion

It was concluded from the findings that treatment with PGF₂α on Day 15 post partum had a beneficial effect on the reproductive performance in cows were treated with M₁ and M₂ protocol. Furthermore, administration of a GnRH agonist on Day 23 after AI did not improve reproductive performance for cows receiving first post partum TAI after Heatsynch protocol.

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