OVARIAN STATUS, SERUM PROGESTERONE (P4) LEVEL AND CONCEPTION RATE IN OVSYNCH PLUS CIDR TREATED POSTPARTUM BUFFALOES*

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ABSTRACT

Postpartum buffaloes (n = 23) not resuming or not showing observed estrus within 60 days after calving were classified based on palpable ovarian structures as ovsynch plus CIDR-subestrus (n = 12) and ovsynch plus CIDR-anestrus (n = 11) groups. Ovsynch plus CIDR buffaloes were subjected to ovsynch protocol which consisted of 100 $\mu$g i.m. injection of GnRH on day 0, 25 mg i.m. injection of PGF$_2\alpha$ on day 7, another 100 $\mu$g i.m. injection of GnRH on day 9 and timed insemination 16 to 18 h after second GnRH injection (d 10) and were inserted with CIDR device for a period of 7 days starting from first GnRH injection (d 0) to PGF$_2\alpha$ injection (d 7) for synchronization of ovulation. Blood samples were collected from all experimental animals on days -10, 0, 7 and 9 for the estimation of progesterone to predict the physiological status of different treatment groups and were combined to produce eight different permutations viz. HHH (high, high, high), LHH, LHL, HHL, LLL, LLH, HLL and HLH. First four groups represented those buffaloes that had elevated progesterone on d 7 (presence of CL) at the time of the PGF$_2\alpha$ injection while the last four groups represented those that had low progesterone at the time of PGF$_2\alpha$ (d 7) injection. The fertility response to ovsynch plus CIDR in the buffaloes of HHL class was found to be higher when compared to other progesterone classes in both treatment groups.

Key words: Ovarian status, serum progesterone, ovsynch plus CIDR, Postpartum, Buffaloes

INTRODUCTION

The major limitation of GnRH and PGF$_2\alpha$ based synchronization protocols was the inability of GnRH to turnover dominant follicles late in the estrous cycle leading to premature estrus in 8 to 10 per cent of treated animals (Geary et al., 2000). Alternatively, the inclusion of an exogenous progestogen such as CIDR during the interval between GnRH and PGF$_2\alpha$ injection prevented premature estrus and increased conception rates (Steckler et al., 2002). Although much work has been done using ovsynch plus CIDR in synchronization of ovulation in cattle, information on their use in buffaloes especially postpartum lactating Murrah

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buffaloes is limited. Hence the present study was undertaken to compare the effect of ovarian status, serum progesterone level and conception rate in ovsynch plus CIDR treated buffaloes.

**MATERIALS AND METHODS**

Twenty three apparently healthy lactating buffaloes maintained at Central Cattle Breeding Farm, Alamathi, with a history of absence or presence of estrus signs within 60 days post partum and/or palpable ovarian structures during rectal examination were selected and divided into ovsynch plus CIDR -subestrus and ovsynch plus CIDR – anestrus groups. Serum progesterone (P4) levels were estimated on day 10 and day 0 to confirm the presence or absence of cyclic activity of the selected buffaloes. The ovsynch subestrus and anestrus group buffaloes were injected with 100 μg GnRH (Gonadorelin Acetate, Fertiline, Vetoquinol, Canada) i.m. on the day of start of treatment on day 0, 25 mg PGF2α, i.m. (Dinoprost tromethamin, Lytalyseâ, Pharmacia, Belgium) on day 7, another 100 μg GnRH i.m. on day 9 and timed insemination 16 to 18 h after the second GnRH injection on day 10. Buffaloes were subjected to ovsynch plus CIDR protocol which included intra vaginal placement of CIDR device for a period of 7 days starting on day 0 to day 7 for synchronization of ovulation. The experimental buffaloes were observed frequently for estrus signs during the period of treatment and after PGF2α injection. Blood samples were collected from all animals on d -10, d 0, d 7 and d 9 for the estimation of serum progesterone using solid-phase Radio Immuno Assay technique with the help of P4 kits (Coat-A-Count, Diagnostic Products Corporation, USA). Serum samples collected on d 0, d 7 and d 9 from each animal were combined to produce eight permutations to predict the physiological status viz. HHH (high, high, high), LHH, LHL, HHL, LLL, LLH, HLL and HLH and the first four groups represented the buffaloes that had elevated P₄ on d 7 (presence of CL) at the time of the PGF₉α injection while the last four groups represented those that had low P₄ at the time of PGF₉α injection. First service conception rate was calculated as percentage of animals that conceived to fixed time insemination at induced estrus in each group.

**RESULTS AND DISCUSSION**

The incidence of cyclicity and conception rate of buffaloes based on concentration of P₄ in each of the serum samples collected on d 0, 7 and 9 in ovsynch treated postpartum subestrus and anestrus buffalo is presented in the Table. Of the 23 ovsynch buffaloes assigned to a P₄ class, 95.65 (22/ 23) per cent had high P₄ at d 7 with 1 HHH, 1 LHH, 11 LHL and 9 HHL buffaloes.

In the present study, one HHH subestrus buffalo of ovsynch group did not conceive. The HHH buffalo was likely in diestrus or in early proestrus at the time of the first GnRH injection and had failed to undergo luteolysis because their GnRH induced corpus luteum was not responsive to PGF₉α (Wiltbank et al., 1995) and failed to completely regress (Moreira et al., 2000) resulting in high P₄ at d 9. It is also possible that some cows receiving ovsynch ovulated smaller follicles due to administration of GnRH when compared with cows ovulating after a spontaneous estrus after acquisition of ovulatory capacity, but before the final stages of follicular maturity. Ovulation of smaller follicles might have resulted in smaller corpus luteum that produced less P₄ (Vasconcelos et al., 1999) and perhaps exhibited a delayed responsiveness to PGF₉α due to expression of fewer PGF₉α receptors. Incomplete luteal regression had been reported in lactating dairy cows receiving ovsynch, with 7 per cent of cows exhibiting high (> 2.0 ng/ml) P₄, 48 h after PGF₉α (Moreira et al., 2001). Furthermore, MacMillan and Henderson (1984) reported that 8 per cent of lactating dairy cows failed
### Table

Ovarian status, serum progesterone (p4) level and conception rate in ovsynch plus CIDR treated postpartum buffaloes

<table>
<thead>
<tr>
<th>Progesterone</th>
<th>Physiological Status</th>
<th>Ovsynch plus CIDR (n=23)</th>
<th></th>
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</thead>
<tbody>
<tr>
<td></td>
<td></td>
<td>Incidence of Cyclicity per cent (no./no.)</td>
<td>Conception Rate per cent (no./no.)</td>
</tr>
<tr>
<td></td>
<td>Sub-estrus</td>
<td>An-estrus</td>
<td>Total</td>
</tr>
<tr>
<td>High progesterone on d 7 at the time of PGF&lt;sub&gt;2α&lt;/sub&gt;</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>HHH</td>
<td>No luteolysis</td>
<td>8.33 (1/12)</td>
<td>0 (0/11)</td>
</tr>
<tr>
<td>LHH</td>
<td>Ovulation after d 0; no luteolysis</td>
<td>0 (0/12)</td>
<td>9.09 (1/11)</td>
</tr>
<tr>
<td>LHL</td>
<td>Synchronized plus luteolysis</td>
<td>16.66 (2/12)</td>
<td>81.81 (9/11)</td>
</tr>
<tr>
<td>HHL</td>
<td>Synchronized plus luteolysis</td>
<td>75.00 (9/12)</td>
<td>0 (0/11)</td>
</tr>
<tr>
<td>Total</td>
<td>100.00 (12/12)</td>
<td>90.90 (10/11)</td>
<td>95.65 (22/23)</td>
</tr>
<tr>
<td>Low progesterone on d 7 at the time of PGF&lt;sub&gt;2α&lt;/sub&gt;</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>LLL</td>
<td>Anestrus</td>
<td>0 (0/12)</td>
<td>9.09 (1/11)</td>
</tr>
<tr>
<td>LLH</td>
<td>Anestrus; spontaneous ovulation</td>
<td>0 (0/12)</td>
<td>0 (0/11)</td>
</tr>
<tr>
<td>HLL</td>
<td>Mid cycle (d 11-16) on d 0</td>
<td>0 (0/12)</td>
<td>0 (0/11)</td>
</tr>
<tr>
<td>HLH</td>
<td>Late cycle (d 17-19) on d 0</td>
<td>0 (0/12)</td>
<td>0 (0/11)</td>
</tr>
<tr>
<td>Total</td>
<td>0 (0/12)</td>
<td>9.09 (1/11)</td>
<td>4.34 (1/23)</td>
</tr>
</tbody>
</table>

One LHH buffalo of the ovsynch plus CIDR group was in anestrus at the time of first GnRH injection. The LHH anestrus buffalo possibly would have responded to the first GnRH injection but, the PGF<sub>2α</sub> induced incomplete luteolysis might have resulted in a persistent follicle (non-ovulatory) (Tugiramungu et al., 1994) that would have
Ovarian status, serum progesterone ....

Ovulated following administration of the second dose of GnRH, due to LH release caused by GnRH (Wiltbank et al., 1996). However, the LHH buffalo in the present study did not conceive. The possible retention of the Graafian follicle for an extended period might have lead to damage of the oocyte to such an extent that even inseminating close to the time of ovulation might not have ensured conception (Duchens et al., 1994).

Nine out of eleven LHL buffaloes of ovsynch plus CIDR were in anoestrus, while the remaining two were in suboestrus. The increased responsiveness of anoestrus buffaloes to ovsynch demonstrated the effectiveness of GnRH for inducing ovulation of anoestrus dairy cows similar to that reported previously (Stevenson et al., 1999). The suboestrus buffaloes of LHL P4 classes might have been in prooestrus at the time of first GnRH injection and possibly would have undergone synchronized follicular and luteal function in response to the ovsynch plus CIDR protocol (Cordoba and Fricke, 2002). Two ovsynch plus CIDR- anoestrus and one ovsynch plus CIDR-suboestrus groups conceived, thus, indicating that exogenous progestin treatment during the interval between GnRH and PGF2α injection prevented premature estrus and increased conception rates of anoestrus animals (Thompson et al., 1999 and Stevenson et al., 2000).

Suboestrus buffaloes exhibiting cycles with two follicular waves and initiating ovsynch during the later stages of oestrus cycle would exhibit the HHL P4 profile and higher conception rates because of the greater likelihood of ovulation in response to the first GnRH injection which would prevent premature ovulation and asynchrony with timed AI (Cordoba and Fricke, 2002). In the present study too, the highest conception rates of 44.44 per cent was observed in ovsynch plus CIDR HHL buffaloes when compared to other P4 classes.

Of the 23 ovsynch plus CIDR buffaloes assigned to a P4 class 4.34 (1/23) per cent had low P4 at the time of PGF2α with 1 LLL buffalo. The LLL buffalo of ovsynch plus CIDR did not conceive possibly due to the fact that the LLL buffalo was anoovular and had failed to ovulate in response to the first GnRH injection.

The results of the present study have shown that ovsynch plus CIDR - suboestrus group responded better with increased conception rates when compared to ovsynch plus CIDR - anoestrus group. Further, fertility response of ovsynch plus CIDR buffaloes of HHL class was found to be higher when compared to other P4 classes in both treatment groups. Thus, to conclude ovsynch plus CIDR protocol may be an ideal strategy to improve fertility in lactating postpartum suboestrus buffaloes than in lactating partum anoestrus buffaloes.

REFERENCES


