Synchronization of Estrus in Naeimi Ewes Following
Treatment with Progestagens or Prostaglandin F₂α

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Abstract :
The efficacy of using progesterone releasing interavaginal device (PRID), PRID without estradiol benzoate, veramix sheep sponge and prostaglandin F₂α in inducing and synchronizing estrus were evaluated in Naeimi ewes. All four treatments were successfully able to synchronize estrus. The interval to induce estrus was significantly (P<0.05) shorter and estrous intensity was significantly higher in PRID-treated group compared to other treatment groups. Serum progesterone values during subsequent estrous cycle and pregnancy rate were similar in all treatment groups.

The results indicated that progestagens and prostaglandin F₂α can effectively be used in management of reproduction in Naeimi sheep.

Introduction :
Inspite of the numerical and economical importance of the Awassi sheep in Saudi Arabia, few planned research investigations were conducted to study the various factors which affecting their reproduction.

The study of reproduction in Awassi breed received considerable attentions in neighbouring countries (Hossamo et al., 1985 in Syria; Eliya et al., 1972 in Iraq; Finci 1975 in Palestine; Khalil et al., 1972 in Lebanon). Similar information on the indigenous Naeimi sheep which is a branch of the Awassi breed of Saudi Arabia is scant.

Estrus synchronization has become a vital instrument in the management of reproduction in domestic farm animals (Motlomelo et al., 2002; Fonesca et al., 2008). Its benefits include a planned breeding programme and a reduction in labor costs in terms of estrus detection and care of the newborn. Estrus synchronization has also become an important tool for embryo transfer.

The objective of this study was to evaluate the efficacy of progestagens and prostaglandin F₂α in synchronizing estrus in Awassi (Naeimi) sheep, and to record pregnancy thereafter.
Materials and Methods:

Animals and treatments:

Eighty mature (aged 2-4 years and weighing 30-40kg) (Naeimi sheep) were used in the study. They were housed in pens under conditions of natural day length and temperature. They were fed on a roughage mixture composed of 50% green fodder (Medicago sativa) and 50% Alfalfa hay offered ad libitum. Water and salt licks was available at all times. All ewes were checked for estrus when they stand for mounting by a ram this is termed day 0 of the estrous cycle. Animals were allocated randomly and equally to groups of 20 animals each:

Group A: ewes were injected with PGF2α (Dinoprost Tromethamine, Upjohn Company, Kalamazoo, USA; 10mg i.m) at day 0 and then at 11-day intervals.

Group B: ewes were treated with progesterone releasing intravaginal device (PRID). The PRID used had a progesterone content of 1.55g, and a capsule containing 10mg of estradiol benzoate attached the coil.

Group C: ewes were treated with PRID after removal of the capsule of estradiol benzoate from to the coil.

Group D: ewes were treated with Veramix sheep sponges (Upjohn Ltd, Fleming Way, Crawley, Sussex, England) containing 60mg 6-methyl-17-acetoxy-progesterone. Sponges and PRIDS were inserted in the anterior vagina at day 0, and were removed 14 days later.

Ewes were exposed three times daily (0600, 1200 and 1800h) post treatment to aproned rams. The duration of standing estrus was notified by immobility when mounted by a male, while the length of the estrous cycle was regarded as the interval from the end of estrus to the beginning of the next one in the same ewe. The intensity of estrus was scored on a scale from zero to 3 as the degree of expression of restlessness, standing to be mounted, vocalization, and swelling of vulva and mucus discharge. Animals were then bred by natural service and pregnancy was recorded.

Collection of Blood Samples:

After completion of one normal cycle, blood sampling was commenced on day 2 of estrous cycle. Jugular blood (5ml) is obtained once daily by direct venepuncture using 23 gauge needles. From day 12 through to day of estrus, samples were taken at two hour intervals between 07:00 and 17:00 hours.
All blood samples were collected into chilled, heparinised tubes, stored in ice for a short period and then centrifuged at 1500g for 10 minutes. Plasma was separated and stored at -30°C until analysis.

**Radioimmunoassay of hormones**

Plasma progesterone was measured by the radioimmunoassay (RIA) method described by Homeida (1986), and Homeida & Al-Eknah (1992). Progesterone antibody was raised in rabbits against Progesterone-11-succiny-bovine serum albumin, and used at a final dilution of 1:7000; cross-reactions were 100% with progesterone and <0.1% with corticosterone, desoxycorticosterone and ketocorticosterone. The intra- and inter-assay coefficients of variation were 5.3% and 9.1%, respectively.

**Statistical analysis**

Comparisons of hormone concentrations over time were made by ANOVA, taking of repeated measures from same animal (Steele and Torrie 1988).

**Results**

Estrual responses following treatment with PRIDs, veramix sponge or PGF$_{2\alpha}$ treatment in Naeimi ewes are presented in table (1). Neither PRIDs nor sponge were lost during the treatment periods. Estrous was synchronized in most of ewes by the four treatment methods. The interval to onset estrus were significantly (P<0.05) shorter in group B (44.2 hour) than in group C (52.6 hour) or group D (77.6 hour) following removal of the devices. The interval in group B was similar to group A (41.3 hour) following the second PGF$_{2\alpha}$ injection. The duration of subsequent estrous cycle was 16.3, 16.5, 16.2 and 17.3 days in groups A, B, C and D, respectively.

The estrus intensity score was significantly (P<0.05) higher in group B compared to other groups.

Number of animals in estrous and number of pregnant animals were higher though not significant in group B compared to other groups.

Mean serum progesterone concentrations are presented in table (2). Values of serum progesterone concentrations were significantly (P<0.05) higher in group B compared to other groups during device insertion. Serum progesterone concentrations increased steadily following first PGF2α treatment or device insertion. The pattern of the hormone was similar during the subsequent estrous cycle but tended to be higher in group B, C and D compared to group A.
### Tabel (1) : Estrual responses following treatment with PRIDs Veramix sponge or PGF$_{2α}$ treatments in Naeimi ewes

<table>
<thead>
<tr>
<th>Parameter</th>
<th>Group A (PGF$_{2α}$ treated)</th>
<th>Group B (PRID Treated)</th>
<th>Group C (PRID with-estradiol treatment)</th>
<th>Group D (Veramix treated)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Number of treated animals</td>
<td>20</td>
<td>20</td>
<td>20</td>
<td>20</td>
</tr>
<tr>
<td>Number of animals lost device</td>
<td>-</td>
<td>0</td>
<td>0</td>
<td>0</td>
</tr>
<tr>
<td>Number in estrus</td>
<td>16</td>
<td>18</td>
<td>14</td>
<td>15</td>
</tr>
<tr>
<td>Mean interval to estrus (hour)</td>
<td>41.3± 2.1</td>
<td>44.2± 1.2</td>
<td>52.6± 1.4*</td>
<td>77.6± 3.1*</td>
</tr>
<tr>
<td>Mean estrus intensity (score 12)</td>
<td>6± 0.1</td>
<td>12± 0.3*</td>
<td>6± 0.2</td>
<td>6± 0.1</td>
</tr>
<tr>
<td>Duration of estrous cycle (days)</td>
<td>16.2± 0.3</td>
<td>16.5± 0.3</td>
<td>16.2± 0.2</td>
<td>17.3± 0.3</td>
</tr>
<tr>
<td>Number of pregnant animals</td>
<td>13</td>
<td>16</td>
<td>12</td>
<td>12</td>
</tr>
</tbody>
</table>

* P<0.05, significantly different from their counterparts.
Table (2)
Mean (± SD) serum progesterone concentration (ng/ml) during PGF$_{2\alpha}$ treatment or vaginal device insertion and removal in Naeimi ewes.

<table>
<thead>
<tr>
<th>Days following device insertion or first PGF$_{2\alpha}$ injection</th>
<th>Group A</th>
<th>Group B</th>
<th>Group C</th>
<th>Group D</th>
</tr>
</thead>
<tbody>
<tr>
<td>2</td>
<td>0.5 ± 0.2</td>
<td>2.1 ± 0.2</td>
<td>1.5 ± 0.2</td>
<td>1.2 ± 0.2</td>
</tr>
<tr>
<td>4</td>
<td>1.1 ± 0.2</td>
<td>4.4 ± 0.3*</td>
<td>2.5 ± 0.2</td>
<td>2.1 ± 0.2</td>
</tr>
<tr>
<td>6</td>
<td>2.4 ± 0.3</td>
<td>6.1 ± 0.4*</td>
<td>3.1 ± 0.2</td>
<td>3.3 ± 0.3</td>
</tr>
<tr>
<td>8</td>
<td>3.9 ± 0.3</td>
<td>6.6 ± 0.4</td>
<td>3.9 ± 0.3</td>
<td>4.2 ± 0.3</td>
</tr>
<tr>
<td>10</td>
<td>4.2 ± 0.3</td>
<td>7.4 ± 0.4*</td>
<td>4.1 ± 0.3</td>
<td>5.1 ± 0.3</td>
</tr>
<tr>
<td>12</td>
<td>2.1 ± 0.2</td>
<td>7.3 ± 0.4*</td>
<td>4.1 ± 0.3</td>
<td>5.3 ± 0.4</td>
</tr>
<tr>
<td>14</td>
<td>7.1 ± 0.3*</td>
<td>4.6 ± 0.3</td>
<td>5.5 ± 0.4</td>
<td></td>
</tr>
</tbody>
</table>

<table>
<thead>
<tr>
<th>Days following device removal or second PGF$_{2\alpha}$ injection 2</th>
<th>Group A</th>
<th>Group B</th>
<th>Group C</th>
<th>Group D</th>
</tr>
</thead>
<tbody>
<tr>
<td>4</td>
<td>0.4 ± 0.1</td>
<td>0.5 ± 0.1</td>
<td>0.4 ± 0.1</td>
<td>0.3 ± 0.1</td>
</tr>
<tr>
<td>6</td>
<td>0.4 ± 0.1</td>
<td>1.1 ± 0.1</td>
<td>1.4 ± 0.2</td>
<td>1.2 ± 0.1</td>
</tr>
<tr>
<td>8</td>
<td>0.25 ± 0.2</td>
<td>2.4 ± 0.2</td>
<td>2.6 ± 0.2</td>
<td>2.5 ± 0.2</td>
</tr>
<tr>
<td>10</td>
<td>1.1 ± 0.3</td>
<td>4.5 ± 0.3</td>
<td>5.6 ± 0.3</td>
<td>5.4 ± 0.3</td>
</tr>
<tr>
<td>12</td>
<td>4.3 ± 0.3</td>
<td>5.2 ± 0.3</td>
<td>5.3 ± 0.3</td>
<td>5.1 ± 0.4</td>
</tr>
<tr>
<td>14</td>
<td>3.6 ± 0.3</td>
<td>3.2 ± 0.3</td>
<td>3.1 ± 0.3</td>
<td>2.9 ± 0.3</td>
</tr>
<tr>
<td>16</td>
<td>2.3 ± 0.2</td>
<td>2.1 ± 0.2</td>
<td>2.4 ± 0.2</td>
<td>2.3 ± 0.2</td>
</tr>
<tr>
<td>18</td>
<td>0.6 ± 0.1</td>
<td>0.5 ± 0.1</td>
<td>6.4 ± 0.1</td>
<td>0.3 ± 0.1</td>
</tr>
</tbody>
</table>

* P< 0.05
Discussion:

The four estrus synchronizing methods have effectively synchronized estrus in Naeimi sheep. Comparable efficacy achieved by progestagens in synchronizing estrus has been reported in Syrian Awassi ewes (Zarkawi et al. 1999) and other breeds of sheep (kusakari et al., 1991; Das et al. 2001; Mutiga and Mukasa – Mugerwa 1992) and goats (Fonseca et al. 2008). The mean interval to estrous was shorter and the intensity of estrus was higher in animals treated with PRID than in other animal groups. This is probably due to estrogen being incorporated in the PRID. Enhanced estrus expression and behavior was noticed when estradiol was given in conjunction with PRID in cows (Walton and king 1984, Narasimha & suryapakasam 1991)

Furthermore, (coetzer et al., 1988) showed that removal of estradiol benzoate from a PRID device resulted in poor estrus response.

The degree of synchrony and interval to estrus following the second PGF2α injection were consistent with earlier reports in different breeds of sheep (Acritopoulou et al., 1978; Haesign 1978; Oydiji et al., 1990).

Although fertility trials were not assessed critically in this experiment but pregnancy rate was similar with all synchronized methods. Pregnancy rate was high in sheep following progestagen (Das et al., 2004) and prostagland via F2α (Gardenas et al. 1993) treatments.

Administration of PRID resulted in significantly higher progesterone concentration compared to PRID without estradiol benzoate or veramix sponge. This is probably due to modulating effect of estradiol on the absorption of progesterone from vagina in the PRID group and the difference in physicochemical properties between the natural progesterone contained in PRID and the synthetic progesterone contained in veramix sponge.

The pattern of progesterone following the device removal or following the second PGF2α injection were similar to the natural estrous cycle in sheep and cattle (Cardenas et al., 1993; Broadbent et al., 1993).

In conclusion, the present study indicated that intravaginal progestagen device or prostaglandin F2α can be used effectively for induction and synchronization of estrus in Naeimi sheep. Behavioral estrus was better expressed in the PRID group. Pregnancy rate was similar in all treatment methods.

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References:


الملخص:

لقد تم تقييم مقدرة أحداث تزامن الشبق في النجع النعيمي بواسطة البروجستاجين F_{2a} 
البروجستاجين أو الحشوة البروجستاجينية بدون الاستروجين أو الحشوة الأسفنجية 
الدوموسك أو البروجستاجينات - ف - الفا . لقد تم أحداث تزامن الشبق بنجاح 
باستخدام الدواء المستخدمة . لقد تم أحداث الشبق في وقت اقتصر عادة وأن ظواهر 
الشبق مكان بصورة أوضح في النجع المعالجة بالحشوة البروجستاجينية بالمقارنة مع بقية 
المعالجات ولقد عُدّت ترطيب البروجستاجينات في الدورة اللاحقة للمعالجة والحمل 
متشابه في المعالجات الأربعة . هذه النتائج تشير إلى مقدرة البروجستاجينات 
والبروجستالاندين في إدارة النواحي التناسلية في نجع النعيمي.