

## A COMPARATIVE STUDY OF ESTRUS INDUCTION WITH LONG-TERM AND SHORT-TERM PROGESTERONE IN EWES DURING NON-BREEDING AND BREEDING SEASONS

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### SUMMARY

The objective of this trial was to compare the efficacy of long-term and short-term progesterone treatments on induction of estrus and pregnancy rate in Rahmani ewes during non-breeding and breeding seasons. Twenty-four animals were randomly grouped for two treatments with intravaginal progesterone device: Group A) during breeding season and Group B) during non-breeding season. Furthermore, both groups were subdivided into two treatments: A1 and B1 for short-treatment (7 days) and A2 and B2 for long-treatment (12 days). The progesterone treatment was a vaginal sponge containing 40 mg medroxy progesterone acetate (MAP). Equine chorionic gonadotrophin (eCG) and cloprostenol (PGF $2\alpha$ ) were intramuscularly administered to all ewes at the time of sponge removal. After the detection of estrus, the animals were hand-mated naturally.

During the breeding season, mean percentages of estrus and pregnancy were 100% and 83.33%, respectively in both the short-term and long-term treated groups. During the non-breeding season, the average values of estrus were 100% in short-term and 100% for long-term progesterone treatment, but the pregnancy rates were 66.67 and 50.00% in the short- and long-term treated groups, respectively.

The treatment with progesterone for short-term (7 days) was effective to induce estrus in sheep during both breeding and non-breeding seasons.

### **Keywords:**

Progesterin - Breeding and non-breeding seasons - Rahmani Egyptian ewes.

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## INTRODUCTION

Most sheep breeds in Egypt are seasonal polyestrous animals. The estrus season (breeding season) starts in autumn and lasts until spring and is connected with reduction in the light intensity (Tempest and Minter, 1987).

Estrus synchronization has been the main method used to manage reproduction in farm animals. Progestagen and prostaglandin analogues are widely used in sheep for estrus synchronization (Beck et al., 1993). Intravaginal sponges containing progesterone are one of the most applied treatments for estrus synchronization in small ruminants during the breeding and non-breeding seasons. Intravaginal sponges are usually inserted over periods of 12 to 14 days and used together with equine Chorionic Gonadotrophin (eCG), particularly out of season, administered at the time of sponge withdrawal or 48 hours prior to sponge removal (Jainudeen et al., 2000).

Prostaglandin-based estrus synchronization systems control the estrous cycle by terminating the luteal phase through regression of the corpus luteum. This approach is only applicable in cyclic female and hence, prostaglandin-based systems can only be used during the breeding season. Because not all stages of the estrous cycle are similarly receptive to prostaglandin treatment, a double injection system with an interval of 11 days is the most widely used approach in small rumi-

nants (Jainudeen et al., 2000).

Pregnancy rates of progestagen-synchronized ewes were lower during non-breeding than during the breeding season (Crosby et al., 1991). Fertility was related positively to concentrations of progesterone during the treatment (Johnson et al., 1996), probably as a result of more appropriate patterns of follicular development (Johnson et al., 1996), and timing of the LH surge in relation to the onset of estrus. So, the release of progesterone from the sponges declines over time (Van Cleef et al., 1998). Therefore, a short term treatment provides higher average concentrations of progesterone during the treatment period. Such treatments (5-8 days) have shown to be effective during anestrus season (Rodriguez-iglesias et al., 1996) and breeding season (Özturkler et al., 2003) in sheep. Despite the better conception rate obtained, the efficiency of short progesterone priming for estrous synchronization during the breeding season remains limited due to presence of a still functional corpus luteum (Vinoles et al., 2001).

Because ovulation rate and prolificacy are the lowest during the spring/summer period (Hall et al., 1986), gonadotropin treatment at the end of progesterone treatment has been used to increase prolificacy of anoestrous ewes in non-breeding time (Safranski et al., 1992).

Although the idea of inducing and synchronizing breeding activity in anestrus ewes is not new,

data on the treatment of ewes with short-term and long-term progestagen combined with eCG and PGF2 $\alpha$  are few.

The objective of this study was to compare the efficacy of the length of a progestagen treatment (7 days Vs 12 days ) on synchronizing efficiency and pregnancy rate using MAP treatment with eCG and PGF2 $\alpha$  administration in Rahmani ewes in the breeding and non-breeding seasons.

## **MATERIAL AND METHODS**

### **Animals**

A total of 24 non-lactating mature ewes of Rahmani breed aging 18-24 months with a weight of 50-60 kg .Two intact Rahmani rams aging 2-3 years with a known fertility were used in this experiment.

The animals were kept in a sheltered outdoor barn under natural lighting conditions, and kept indoor at night. The animals were dewarmed and their health was checked by blood analysis before starting the experiment. This study was conducted on 60 to 90 days postpartum ewes during the breeding (September - March) and non-breeding (April - June) seasons on a farm in El-Behera province. During the non-breeding period, the proportion of ewes having spontaneous cycling activity (plasma progesterone levels > 0.5 ng/ml) was 13.33%. The animals were managed on green foods mainly of barseem in addition to

wheat straw supplemented with crushed yellow maize. Water and mineral licks were available ad libitum.

### **Methods**

The experiment was performed on two groups of animals: Group A) during the breeding season, Group B) during the non-breeding season. Furthermore, both groups were subdivided into two treatments, A1 , B1 for short-term (7 days) and A2 , B2 for long-term (12 days) progesterone treatments.

### **Treatment**

A vaginal sponge containing 40 mg Medroxy Progesterone Acetate (MAP, sponges, Hand - made) was inserted into the vagina of the ewes for 7 days as a short-term and for 12 days as a long-term progesterone treatment during the breeding and non-breeding seasons (Simonetti et al., 2000).

At the time of sponges withdrawal, all animals received both an intramuscular injection of 500 $\mu$ g cloprostenol sodium (Estrumate, Intervet, Holland) an analogue of PGF2 $\alpha$  , and of eCG (500 IU folligon, Intervet, Holland). After the eCG administrations, the estrous cycle of each animal was checked twice every 12-hour interval using a teaser ram for 4-5 days. After the detection of estrus, the animals were hand-mated naturally (Rodes and Nathanielsz , 1998).

Ewes were considered in estrus when they allowed by the male to mount. Estrus duration was defined as the time elapsed between the first and last accepted mount within the same estrous period.

### Pregnancy diagnosis

Pregnancy was diagnosed using ultrasonography with a transrectal probe 30 days after mating. The number of embryos of each ewe was determined using the transrectal ultrasonography on day 30 and 36 as described by Schrick and Inskip (1993).

### The following traits were evaluated:

#### Estrus response:

(number of ewes showing estrus behavior/ total treated ewes in each group) x100.

#### Pregnancy rate:

(number of ewes pregnant / number of ewes showing estrus in each group) x100.

#### Statistical analysis

Data were analyzed with chi-square analysis to

compare the estrus and pregnancy rates among the groups and a t-test was performed to compare the mean time of occurrence of estrus after sponge removal (Snedecor and Cochran,1980).

## RESULTS

None of the treated ewes lost its device during the insertion period.

Estrus was observed in 12 out of 12 ewes in the group A (100% ) and also in all animals in group B (100% ) indicating the effectiveness of MPA sponges administered for 7 or 14 days in conjunction with eCG and PGF2 $\alpha$  (Table,1) during the breeding and non-breeding seasons.

The mean for the time of estrus onset was about 42hr for short-term and also 42hr for long-term progesterone treatment in animals of group A1 and A2, respectively (Ttable ,1).

The animals in group B1 and B2 had a mean sponge's withdrawal-estrus 45 and 44 hr, respectively (table, 1).

**Table (1):** Reproductive responses of Rahmani ewes to estrus synchronization during the breeding and non-breeding seasons

Parameters	Breeding season		Non-breeding season	
	Short- term A1 (n=6)	Long- term A2 (n=6)	Short- term B1 (n=6)	Long- term B2 (n=6)
No. of ewes showed estrus (%)	6 (100)	6 (100)	6 (100)	6 (100)
No. of ewes pregnant (%)	5 (83.33)a	5 (83.33)a	4 (66.67)b	3 (50.00)c
Sponge withdrawal – estrus interval (hr)	42.33 $\pm$ 1.5a	41.73 $\pm$ 1.2a	45.12 $\pm$ 1.7a	44.33 $\pm$ 1.45a

Mean  $\pm$  S.E

Means with different superscripts are significantly different ( $p < 0.05$ )

A majority of the ewes came into estrus between 24 and 48 hr following sponge removal (Fig. 1) in group A during the breeding season. Most of the animals in group B exhibited estrus between 36 and 60 hr after sponge's withdrawal (Fig.2).

During the breeding season estrus response and pregnancy rate were the same (100% and 83.33%) in both the short- and long-term progesterone groups, respectively (table, 1). Also, during the breeding season, no significant differences

between the short- and long-term progesterone groups in the sponge withdrawal time (42.33 vs.41.73 hr ).

During the non-breeding season, mean of estrus and pregnancy rates were 100% and 66.67% in the short-term group, and 100% and 50% in the long-term group, respectively. There were no statistically significant differences between the short-term group and the long-term group for these reproductive parameters (Table, 1).

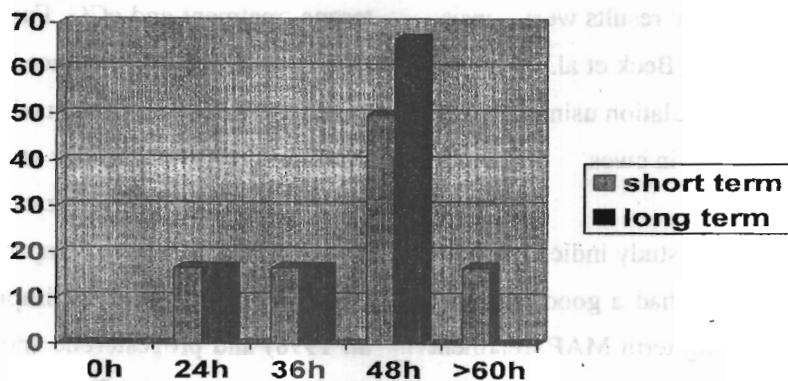


Fig.1 Distribution of estrus over the 2.5 days after inserts withdrawal during breeding season.

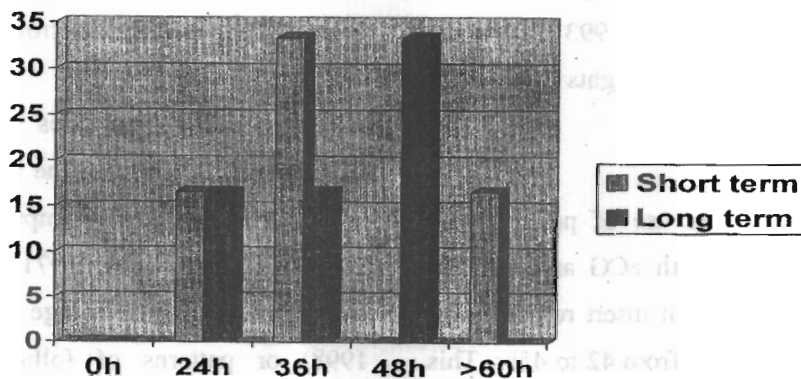


Fig.2 Distribution of estrus over the 2.5 days after inserts withdrawal during non breeding season.

## DISCUSSION

The use of eCG in conjunction with intravaginal progesterone treatment, regardless of PGF2 $\alpha$  administration, was found to be efficient methods for estrus induction and synchronization in the ewes during the non-breeding season (Dogan and Nur,2006). In the current work work, the effect of treatment on estrous expression was evident when eCG and PGF2 $\alpha$  were incorporated into the synchronization protocol which occurred in 100% of ewes during the non-breeding and breeding seasons. These results were consistent with a previous report (Beck et al., 1996) on synchronized estrus and ovulation using Progestagen with eCG and PGF2 $\alpha$  in ewes.

Results obtained in current study indicated that a short term MAP treatment had a good efficacy compared to that of a long-term MAP treatment during both the breeding and the non-breeding seasons. The estrus response to short-term MAP treatment was comparable to that previously reported by others (Beck et al.,1993 ; Beck et al.,1996; Daniel et al., 2001; Knights et al., 2001 and Ozturkler et al.,2003 ).

In the present study, the use of progesterone sponge in combination with eCG and PGF2 $\alpha$  induced an interval between insert removal and the onset of estrus ranging from 42 to 45hr. This time was shorter and less variable than that reported by others (Knights et al., 2001 and

Özturkler et al., 2003). Injection of PGF $\alpha$  th day before sponge withdrawal may prevent the late occurrence of estrus resulting from spontaneous luteolysis of corpus luteum in cycling ewes.

The results obtained with the short-term treatment indicated that this method gave a high level of estrus synchronization and fertility. This result is in agreement with Özturkler et al. (2003) who obtained a synchronization rate of 93.3% using a treatment associating short term progesterone treatment and eCG. Furthermore, the level of synchronization produced by this treatment was similar to that obtained for the 11-days PGF2 $\alpha$  regime (Ozturkler et al.(2003). Moreover , the level of synchronization in breeding and non-breeding groups is comparable with that obtained using Progestagen implant (Cardwell et al. 1998) and progesterone sponges (Beck et al. 1993; Ungerfeld and Rubianes,1999 and Simonetti et al. 2000 ). However, no direct comparison can be made in the present trial because there was no untreated control group.

Improved fertility in ewes synchronized with Progestagen as flurogestone acetate (FGA) and eCG is probably due to improved sperm transport (Hawk and Conley, 1971), synchrony of onset of estrus and LH surge (Van Cleef et al., 1998) or patterns of follicular development (Johnson, et al., 1996).

Response to MAP combined with eCG in ewes during non-breeding season is similar to that obtained with injectable Progestagen (Bachoo and Wani , 1991), progesterone impregnated pessaries and controlled internal drug devices combined with eCG (Daniel et al.,2001) or controlled internal drug devices combined with FSH (Knights, et al. 2001). The conception rate of progesterone-treated ewes was comparable to that previously observed during the breeding (Beck et al., 1993 and Özturkler et al.,2003) and the non-breeding seasons (Zarkawi et al., 1999; Kaya ,et al.,1998 and ; Kridli et al., 2003). The lower fertility of ewes synchronized with Progestagen- eCG treatment during the non breeding season has been attributed to many factors such as impairment of luteinizing hormone secretion, sperm transport and fertilization (Tempest and Minter ,1987). Seasonal effects on reproduction of progestagen-eCG treated ewes appear to be mediated through pituitary gonadotropin secretion with breed differences as to the time and/or intensity of seasonal effects (Ehterikap, 1982).

The current study showed that the use of short-term progesterone treatment was effective to synchronize estrus in breeding and non breeding season in Rahmani sheep. This protocol has some advantages, the duration of synchronization procedure was shorter than that required for other method such as long-term progesterone treatments, progesterone implant or double doses of PGF2 $\alpha$ .

In conclusion, the results from the study herein indicated that long- and short-term, progestagen treatments in combination with eCG and PGF2 $\alpha$  resulted in similar pregnancy rate during breeding season and much pregnancy in short-term than long-term progesterone treatment during the non-breeding season.

Therefore, short-term progestagen treatment can be efficiently used as an alternative to long-term progestagen treatment in Rahmani sheep during non-breeding and breeding seasons.

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## دراسة مقارنة لإستحداث الشبق بالبروجستينات طويلة وقصيرة المدى في الاغنام أثناء موسمي التزاوج وعدم التزاوج .

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تهدف هذه المحاولة لمقارنة كفاءة الحقن بالبروجستينات طويلة وقصيرة المدى في استحداث الشبق ومعدل الحمل في الاغنام أثناء موسمي التزاوج وعدم التزاوج .

استخدم في البحث أربعة وعشرون نعجة ، قسمت عشوائياً للعلاج في مجموعتين باستخدام اللبوس المهبلي :  
مجموعة A ( أثناء موسم التزاوج ) ، مجموعة B ( أثناء موسم عدم التزاوج ) ، ثم قسمت كل مجموعة إلى

قسمين أحدهما للعلاج بالبروجستينات القصيرة المدى لمدة سبعة أيام ( A1 . B1 ) ، والأخرى بالبروجستينات طويلة المدى لمدة ١٢ يوم ( A2 . B2 ) ، وقد كانت اللبوس المهبلية المستخدمة في المجموعتين تحتوي على ٤٠مجم من مادة الميديوروكس بروجستيرون أسيتات مع حقن كلاً من المصل المشمي للأفراس والبروستاجلاندين (كلوربروسيتول) أثناء إزالة اللبوس المهبلي من كل الحيوانات . وبعد إكتشاف ظهور علامات الشباع الجنسي على الاغنام ثم إدخال الذكور إليها لإتمام التلقيح الطبيعي حيث وصلت نسبة حدوث الشباع الجنسي ومعدل الحمل في موسم التزاوج إلى ١٠٠% و ٨٣,٣% في موسم التزاوج على الترتيب بغض النظر عن المدة المستخدم فيها اللبوس المهبلي . أما أثناء موسم عدم التزاوج فقد وصلت نسبة حدوث الشبوع الجنسي إلى ١٠٠% وأما معدل الحمل فقد اختلفت نسبة حدوثه بناءً على الوقت قبل إدخال اللبوس المهبلي حيث كانت ٦٦,٦٧% ، ٥٠% في القصير والطويل المدى على الترتيب . وعلى ذلك فقد تبين أن العلاج باللبوس المهبلي لفترة قصيرة ( ٧ أيام ) أحدث نجاحاً في إحداث الشبق في الاغنام الرحماني سواء في موسمي التزاوج وعدم التزاوج .