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## Medroxyprogesterone priming and response to the ram effect in Corriedale ewes during the nonbreeding season

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### Abstract

The “ram effect” (RE) is an inexpensive technique that allows farmers to obtain out-of-season lambs. Five hundred and ninety-six Corriedale ewes were used in three experiments to determine the effectiveness of different medroxyprogesterone (MAP) treatments associated with the ram effect during the nonbreeding season. The aim of the first experiment was to evaluate the effectiveness of short-term (6-day) MAP priming. We obtained similar results in estrus incidence and fertility after using MAP sponges for 6, 9, and 13 days. In the second experiment, we compared the effect of sponges containing 20, 40, or 60 mg of MAP used in 6-day priming. Estrous behavior and fertility were not affected by dosage. In the third experiment, 2.5 mg of MAP was administered in single treatments 0, 1, 3, or 5 days before the introduction of the rams. Medroxyprogesterone administration 1, 3, or 5 days before the introduction of the rams concentrated estrus in ewes 17 to 20 days later. © 2002 Elsevier Science Inc. All rights reserved.

*Keywords:* Estrus induction; Ram stimulus; Seasonal anestrus; Sheep

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### 1. Introduction

The “ram effect” (RE) is an inexpensive technique which is used for induction of estrus during the nonbreeding season in extensive sheep management systems. Many experiments have been conducted previously, mainly in Merino ewes, to study the response of previously

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isolated anestrus ewes to the introduction of rams [1]. Briefly, immediately after rams are introduced, the reproductive system of the ewes responds with an increase in luteinizing hormone (LH) pulsatility and an LH surge. A silent ovulation occurs, without signs of heat. When the corpus luteum (or corpora lutea) regresses after the first ovulation, another ovulation accompanied by estrus occurs 17 to 19 days after introduction of the rams. However, in some ewes, the corpus luteum regresses after 4 to 5 days and another silent ovulation takes place. After this ovulation, a corpus luteum of normal life span is formed, followed by heat 21 to 25 days after the first introduction of the rams [1]. To obtain estrus in connection with the first ovulation, progestogen priming should be applied before rams are introduced [2].

The use of progestogen priming also prevents the occurrence of short luteal phases (SLPs). Twenty milligrams of progesterone administered in a single treatment when rams are introduced has been reported to result in a peak in estrous behavior 18 to 20 days after introduction of the rams [3,4]. If progesterone is substituted with a single dose of 2.5 mg of medroxyprogesterone (MAP), the estrus peak is delayed by at least 72 h [5].

Traditionally, progestogens have been applied in anestrus ewes for 12 to 14 days to mimic a luteal phase. However, short-term (6-day) priming is at least as effective as is traditional priming for estrus induction with equine chorionic gonadotropin (eCG) [6]. The progestogen content of intravaginal devices appears to be critical to obtaining good fertility [7]. When fluorogestone [8] and progesterone [6] content is low, fertility is affected. Results obtained with MAP are somewhat different. Results produced with intravaginal sponges with a lower MAP content than commercial sponges (60 mg) have been reported to be similar. Fertility has been reported to be unaffected by dosage, both in cyclic ewes [9] and in out-of-season ewes [10–12]. This may be explained by the low amounts of MAP that are absorbed compared with that remaining in the sponges at the end of the treatment [13,14].

The purpose of the current study was to determine the effectiveness of different primings with MAP and the subsequent response to the RE. The objective of the first experiment was to measure the effectiveness of intravaginal sponges on estrous behavior and fertility when exposure to MAP was reduced from 14 to 10 and 6 days. The second experiment using sponges with 20, 40, or 60 mg of MAP for 6 days aimed to establish whether MAP dosage would affect estrous behavior and fertility. The purpose of the third experiment was to elucidate the response, with regard to estrous behavior and fertility, to a single dose of MAP given 0, 1, 3, or 5 days before ram introduction into the flock.

## 2. Materials and methods

### 2.1. Animals and management

Three experiments were conducted on a total of 596 Corriedale ewes. Experiments 1 and 2 were performed on the same farm in Colonia, Uruguay (35° S), and Experiment 3 was conducted on a farm located near Diego Lamas, Artigas, Uruguay (31° S), during the nonbreeding season (November to December; natural light (L):dark (D) ratio = 14 h L:10 h D). Ewes were isolated (sight, sound, smell) from rams (minimum distance: 1000 m)

on Day –30 (Day 0 = introduction of the rams). During the experimental period, ewes were grazed on native pastures.

## 2.2. Experiment 1

One hundred and ninety-eight multiparous ewes, weighing  $52.7 \pm 1.2$  kg (mean  $\pm$  standard deviation (S.D.)) and with a body condition score (BCS, where 1 = extremely emaciated, and 5 = excessively fat) of  $3.4 \pm 0.4$  were used. The ewes had lambed more than 5 months previously and all lambs were withdrawn at least 1 month before the experiment started. Ewes were tagged and divided into four homogenous groups, according to BCS. Intravaginal sponges containing 60 mg of MAP (Syntex, Buenos Aires, Argentina) were inserted on Day –14 (Group 14D,  $n = 43$ ), Day –10 (Group 10D,  $n = 48$ ), and Day –6 (Group 6D,  $n = 48$ ). Fifty-nine untreated ewes served as a control group (Group C1). At sponge withdrawal (November 1), all ewes were placed together with 14 sexually experienced marking Corriedale rams. It has previously been reported that a higher proportion of Corriedale ewes ovulate in response to the RE when ewes in estrus are introduced together with the rams [15]. To ensure the availability of ewes in estrus at the start of the experimental period, 36 additional ewes were, therefore, brought into estrus between Day 0 and Day 3 by i.m. administration of 350 IU of eCG (Novormón, Syntex, Buenos Aires, Argentina) after a 6-day 60 mg MAP priming period and introduced into the flock together with the rams.

Sexual receptivity was estimated from marks on the rumps of the ewes twice daily from Day 0 to Day 5 and again from Day 17 to Day 28, and once daily on Days 8, 11, and 14. All ewes were managed together until marked by the rams. Marked ewes were taken out of the flock and rams were removed as necessary to maintain the ram:ewe ratio. Onset of estrus was considered to be between the last observation at which a ewe was unmarked and the time when marking by a ram was first observed.

To determine pregnancy status, transrectal ultrasonography was performed using a Pie Medical 480 equipment with a dual linear 5/7.5 MHz probe (Pie Medical, Maastricht, The Netherlands) on Day 40 in ewes that were mated on Days 1 to 5. A second ultrasonographic examination was performed on Day 60 in ewes that were first mated after Day 17.

## 2.3. Experiment 2

Two hundred and seven multiparous ewes, weighing  $54.4 \pm 1.1$  kg and with a BCS of  $3.5 \pm 0.5$  were used. Ewes were tagged and divided into four homogenous groups, according to BCS.

On Day –6, intravaginal sponges containing either 20 mg (Group L,  $n = 46$ ), 40 mg (Group M,  $n = 47$ ), or 60 mg of MAP (Group H,  $n = 48$ ) were inserted. Sixty-six untreated ewes served as controls (Group C2). Sponges remained in situ for 6 days. At sponge withdrawal (November 14), all ewes were placed with 17 sexually experienced marking Corriedale rams and 50 additional ewes brought into estrus between Day –1 and Day 4 by i.m. administration of 400 IU eCG (Novormón) after a 6–12-day 60 mg MAP priming period.

Sexual receptivity was determined twice daily from Day 0 to Day 7 and from Day 17 to Day 25, and every second day from Day 8 to Day 16. Ewes were managed as in Experiment

1; and the onset of estrus was estimated as in Experiment 1. Using ultrasonography as in Experiment 1, pregnancy was determined on Day 36 in ewes that were mated between Day 1 and Day 7.

Blood was collected from the jugular vein by venipuncture each 24 to 48 h from Day 0 to the onset of estrus. Samples were allowed to clot for 1 h at room temperature and centrifuged for 10–20 min. Serum was kept at  $-20^{\circ}\text{C}$  until progesterone levels were measured.

#### 2.4. Progesterone radioimmunoassay

Progesterone was measured using a direct solid-phase  $^{125}\text{I}$  radioimmunoassay (RIA) (Count-A-Count TKPG, Diagnostic Products Corporation, Los Angeles, CA) with a sensitivity of 0.3 nmol/l. The inter- and intra-assay coefficients of variation were <10%. Results are expressed in nanomoles per liter.

#### 2.5. Definitions

The existence of luteal phases was established according to the following criteria: (1) progesterone concentration exceeded 1.5 nmol/l in two consecutive samples taken 48 h apart; (2) progesterone concentration exceeded 3 nmol/l in one sample when the time to the previous and to the next sample was 48 h. A normal luteal phase (NLP) was defined as a phase with progesterone luteal levels for at least 10 days, and exceeding 6 nmol/l in at least one sample. An SLP was defined as a luteal phase with progesterone luteal levels for at least 48 h and no longer than 4 days.

#### 2.6. Experiment 3

One hundred and ninety-one nulliparous 2-year-old ewes weighing  $37.9 \pm 0.6$  kg and with a BCS of  $3.3 \pm 0.1$  were used for this experiment. Ewes were tagged and divided into five homogenous groups, according to BCS.

Ewes were treated with 2.5 mg of MAP i.m. (Medrosterona, Laboratorio Gador SA, Argentina) on Day  $-5$  (Group D5,  $n = 40$ ), Day  $-3$  (Group D3,  $n = 38$ ), Day  $-1$  (Group D1,  $n = 38$ ), and Day 0 (Group D0,  $n = 37$ ). Thirty-eight untreated ewes served as a control group (Group C3). On Day 0 (November 20), all ewes were placed with 11 sexually experienced Corriedale marking rams and 20 additional ewes brought into estrus between Day 0 and Day 2 by i.m. administration of 320 IU eCG (Novormón) after a 6-day 60 mg MAP priming period.

Sexual receptivity was estimated once daily from Day 0 to Day 5 and from Day 16 to Day 28, and once on Days 8, 11, and 14. Ewes were managed as in Experiment 1; the onset of estrus was estimated as in Experiment 1. Using transrectal ultrasonography as in Experiment 1, pregnancy was determined on Day 58.

#### 2.7. Data presentation and statistical analysis

All results are presented as means  $\pm$  standard error of the mean (S.E.M.), with a significance level of  $\alpha = 5\%$ . Frequencies of ewes in estrus and pregnant ewes were

compared by chi-squared test; individual groups were compared by Fisher's exact probability test. The time from the introduction of rams to the onset of estrus, and to the onset of NLP (Experiment 2) was compared by analysis of variance (ANOVA); regression analysis was performed for the duration of NLP and NLP onset (Experiment 2). Frequency of ewes that responded initially with an NLP or an SLP (an SLP only, or one followed by an NLP) was compared by chi-squared test and individual groups were compared using Fisher's exact probability test (Experiment 2).

### 3. Results

#### 3.1. Experiment 1

Ewes came into estrus in two periods: Days 1 to 5 and Days 17 to 28. Table 1 presents the frequency of ewes in estrus and the mean time to estrus onset. The distribution of estrus in primed groups was similar, irrespective of the duration of priming. Fig. 1 shows the distribution of estrus in Group C1 (controls) and Group 6D, as representative of all primed groups. A greater proportion of ewes from Groups 14D, 10D, and 6D than of ewes from Group C1 came into estrus during the first estrous period ( $P < 0.001$ , Table 1). In the second estrous period, a greater percentage of control ewes than of primed ewes came into estrus ( $P < 0.001$ , Table 1), but the ratio of ewes in estrus:ewes that had not come into estrus present in the flock during that period was similar for all groups (43/47, 12/17, 6/11, and 8/9 for C1, 6D, 10D, and 14D respectively,  $P > 0.05$ ). The total ratio of ewes in estrus:ewes that had not come into estrus was similar for all groups ( $P > 0.05$ , Table 1).

Of 198 ewes in this experiment, 107 (54%) conceived. The conception rate for the 183 ewes which had come into estrus was 58.5% (107/183). Conception rates, which are shown in Fig. 2, were similar between groups in each estrous period. However, the overall conception rate in the second estrous period (49/69, 71.0%) was higher than that in the first estrous period (58/114, 50.9%;  $P < 0.01$ ).

Table 1

Frequency of ewes in estrus (%) on Days 1 to 5 and Days 17 to 28, and time (days) from introduction of rams to onset of estrus (Experiment 1)

Group	Days 1 to 5		Days 17 to 28		Total ewes in estrus, <i>n</i> (%)
	Ewes in estrus, <i>n</i> (%)	Days to estrus onset	Ewes in estrus, <i>n</i> (%)	Days to estrus onset	
C1	12/59 (20.3) <sup>a</sup>	2.6 ± 0.4	43/59 (72.9) <sup>a</sup>	24.3 ± 0.4 <sup>a</sup>	55/59 (93.2)
6D	31/48 (64.6) <sup>b</sup>	2.3 ± 0.1	12/48 (25.6) <sup>b</sup>	19.4 ± 0.4 <sup>b</sup>	43/48 (89.6)
10D	37/48 (77.1) <sup>b</sup>	2.3 ± 0.1	6/148 (12.5) <sup>b</sup>	18.8 ± 0.6 <sup>b</sup>	43/48 (89.6)
14D	34/43 (79.1) <sup>b</sup>	2.3 ± 0.1	8/43 (18.6) <sup>b</sup>	19.1 ± 0.4 <sup>b</sup>	42/43 (97.7)

On Day 0, rams were introduced to previously isolated anestrous Corriedale ewes primed for 6 (Group 6D), 10 (Group 10D), or 14 days (Group 14D) with intravaginal sponges containing 60 mg of medroxyprogesterone. Control ewes (C1) remained untreated. <sup>a</sup> vs. <sup>b</sup>  $P < 0.001$ .

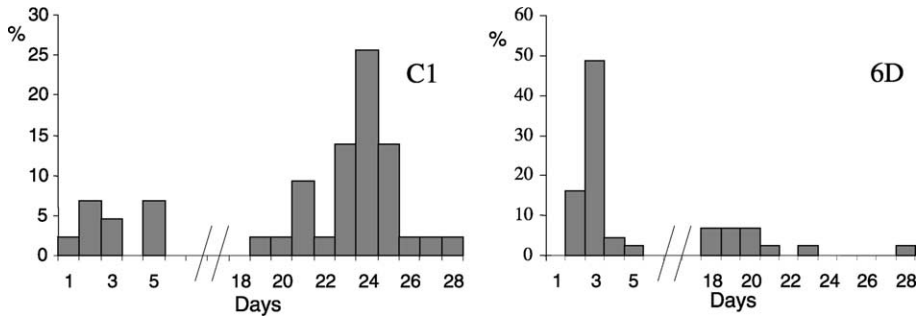


Fig. 1. Onset of estrus after introducing rams (Day 0) to previously isolated anestrus Corriedale ewes. Group C1 did not receive any priming and the distribution of estrus is representative also of the pattern displayed in the control groups of Experiments 2 and 3 (Groups C2 and C3). Group 6D received an intravaginal medroxyprogesterone sponge for 6 days before introduction of the rams, and is representative of the estrus distribution in Groups 14D and 10D (Experiment 1), and L, M, and H (Experiment 2). Note that the scale of the y-axis differs between the graphs. For details of 10D, 14D, L, M, and H treatments, see the text.

3.2. Experiment 2

Ewes came into estrus in two groups: between Day 1 and Day 7, and between Day 17 and Day 25. Table 2 shows the frequency of ewes in estrus and the mean interval between introduction of the rams and onset of estrus. The distribution of estrus was similar between primed groups (L, M, and H), and the pattern was similar to that of the primed groups of Experiment 1 (Fig. 1). In general, a greater percentage of ewes from Groups L, M, and H than of ewes from Group C2 came into estrus during the first estrous period ( $P < 0.001$ ); in the second estrous period, a greater proportion of C2 than of primed animals came into estrus ( $P < 0.01$ , Table 2). However, the ratio of ewes in estrus:ewes that had not come into

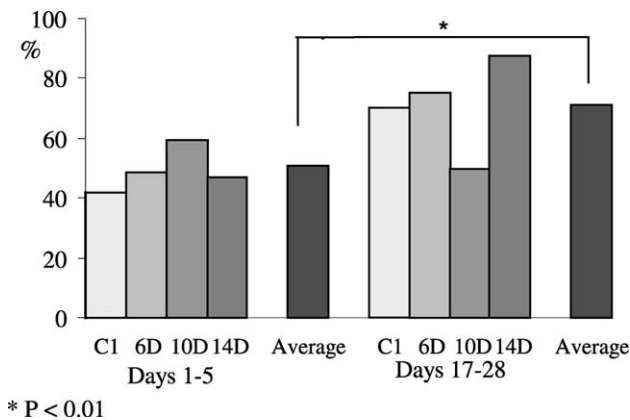


Fig. 2. Experiment 1. Conception rates (%) relating to the periods during which ewes came into estrus. Day 0 rams were introduced to previously isolated anestrus Corriedale ewes primed with an intravaginal MAP sponge for 6 (6D), 10 (10D), or 14 (14D) days. Control (C1) ewes did not receive any priming.

Table 2

Frequency of ewes in estrus (%) on Days 1 to 7 and Days 17 to 25, and time from introduction of rams to onset of estrus (Experiment 2)

Group	Days 1 to 7		Days 17 to 25		Total ewes in estrus, <i>n</i> (%) <sup>c</sup>
	Ewes in estrus, <i>n</i> (%)	Days to estrus onset	Ewes in estrus, <i>n</i> (%)	Days to estrus onset	
C2	7/66 (10.6) <sup>a</sup>	3.6 ± 0.4 <sup>c</sup>	39/66 (59.1) <sup>c</sup>	22.0 ± 0.3 <sup>a</sup>	46/66 (69.7)
L	21/46 (45.7) <sup>b</sup>	2.2 ± 0.2 <sup>d</sup>	15/46 (32.6) <sup>d</sup>	19.1 ± 0.4 <sup>b</sup>	36/46 (78.3)
M	24/47 (51.1) <sup>b</sup>	2.6 ± 0.2 <sup>d</sup>	16/46 (34.0) <sup>d</sup>	19.1 ± 0.4 <sup>b</sup>	40/47 (85.1)
H	25/48 (52.1) <sup>b</sup>	2.5 ± 0.2 <sup>d</sup>	17/48 (35.4) <sup>d</sup>	18.7 ± 0.4 <sup>b</sup>	42/48 (87.5)

On Day 0, rams were introduced to previously isolated anestrous Corriedale ewes primed for 6 days with intravaginal sponges containing 20 mg (Group L), 40 mg (Group M), or 60 mg (Group H) of medroxyprogesterone. Control ewes (C2) remained untreated. <sup>a</sup> vs. <sup>b</sup>  $P < 0.001$ . <sup>c</sup> vs. <sup>d</sup>  $P < 0.01$ . <sup>c</sup>  $P = 0.08$ .

estrus present in the flock during that period was similar for all groups (39/59, 15/25, 16/23, and 17/23 for C2, L, M, and H respectively,  $P > 0.05$ ). When data from the primed groups (Groups L, M, and H) were pooled, a greater percentage of primed (118/141, 83.7%) than of control (46/66, 69.7%) ewes came into estrus ( $P < 0.05$ ). In both periods, primed ewes had an earlier onset of estrus than did control ewes ( $P < 0.01$ ). Conception rates during the first estrous period (i.e. Days 1 to 7) tended to be different between groups (1/7, 11/21, 16/24, and 16/25 for C2, L, M, and H, respectively,  $P = 0.08$ ). The conception rate of the primed groups pooled (43/70, 61.4%) was higher than that of C2 (1/7, 14.3%;  $P < 0.05$ ).

None of the 77 ewes that came into estrus on Days 1 to 7 entered a luteal phase between ram introduction and estrus. Of the remaining 130 ewes, 118 entered the luteal phase before Day 17 (Table 3). A total of 87 came into estrus between Day 17 and Day 25, 39 from Group C2, 15 in Group L, 16 in Group M, and 17 from Group H. The number of ewes

Table 3

Luteal activity in ewes that did not come into estrus in the first estrous period (Days 1 to 7) (Experiment 2)

Group	<i>n</i>	NLP			SLP + NLP		SLP, number (%)
		Number (%) <sup>c</sup>	Ewes in heat on Days 17 to 25	Onset of N L P (days)	Number (%)	Ewes in heat on Days 17 to 25	
C2	51	34/51 (66.7)	28/51 (54.9)	9.3 ± 0.3 <sup>a</sup>	13/51 (25.5)	10/13	4/51 (7.8) <sup>1</sup>
L	23	20/23 (87.0)	13/23 (56.5)	5.6 ± 0.3 <sup>b</sup>	2/23 (8.7)	2/2	1/23 (4.3)
M	22	19/22 (86.4)	15/22 (68.2)	5.6 ± 0.3 <sup>b</sup>	2/22 (9.1)	1/2	1/22 (4.5)
H	22	19/22 (86.4)	17/22 (77.3)	6.3 ± 0.4 <sup>b</sup>	1/22 (4.5)	0/1	2/22 (9.0)

On Day 0, rams were introduced to previously isolated anestrous Corriedale ewes primed for 6 days with intravaginal sponges containing 20 mg (Group L), 40 mg (Group M), or 60 mg (Group H) of medroxyprogesterone. Control ewes (C2) remained untreated. Of 207 ewes, 77 came into estrus on Days 1 to 7. Of the remaining 130, 118 entered a luteal phase before Day 17. Of these 118, the table shows number of ewes with a normal luteal phase (NLP), ewes with a short luteal phase (SLP) followed by an NLP, and ewes with an SLP only, as well as number of ewes that came into heat, and time from introduction of the rams to onset of NLP. <sup>a</sup> vs. <sup>b</sup>  $P < 0.001$ . <sup>c</sup>  $P = 0.08$ ; C2 vs. primed ewes pooled,  $P < 0.01$ .

<sup>1</sup> 1/4 came into estrus.

Table 4

Frequency of ewes that came into estrus (%) on Days 17 to 20 and on Days 21 to 28, and time from introduction of rams to onset of estrus (Experiment 3)

	Ewes in estrus on Days 17 to 20, <i>n</i> (%)	Ewes in estrus on Days 21 to 28, <i>n</i> (%)	Time to estrus onset (days)	Total ewes in estrus, Days 17 to 28, <i>n</i> (%)
C3	11/38 (28.9) <sup>c</sup>	25/38 (65.8) <sup>a</sup>	20.9 ± 0.4 <sup>c</sup>	36/38 (94.7) <sup>g</sup>
D0	1/37 (2.7) <sup>a</sup>	10/37 (27.0) <sup>bc</sup>	23.1 ± 0.7 <sup>d</sup>	11/37 (29.7) <sup>f</sup>
D1	22/38 (57.9) <sup>b</sup>	11/38 (28.9) <sup>b</sup>	18.6 ± 0.4 <sup>c</sup>	33/38 (86.8) <sup>e</sup>
D3	28/38 (73.7) <sup>b</sup>	3/38 (7.9) <sup>d</sup>	17.6 ± 0.3 <sup>ab</sup>	31/38 (81.6) <sup>e</sup>
D5	28/40 (70.0) <sup>b</sup>	4/40 (10.0) <sup>cd</sup>	17.2 ± 0.2 <sup>a</sup>	32/40 (80.0) <sup>e</sup>

On Day 0, rams were introduced to previously isolated anestrous Corriedale ewes previously treated with 2.5 mg medroxyprogesterone 0 (Group D0), 1 (Group D1), 3 (Group D3), or 5 (Group D5) days before the introduction of rams. Control ewes (C3) remained untreated. Different letters within the same column: (a–e)  $P < 0.05$ ; (f, g)  $P < 0.001$ .

responding with an NLP, an SLP followed by an NLP, or an SLP only between Days 2 and 19 is presented in Table 3. The day of onset of NLP influenced the length of the luteal phase ( $r = -0.32$ ,  $P = 0.003$ ).

Of the 12 ewes that did not enter a luteal phase before Day 17, 7 (six from the control group and one from Group M) had progesterone concentrations indicating luteal activity after Day 19. Five ewes did not show any luteal activity during the observation period.

### 3.3. Experiment 3

No ewe was marked by a ram before Day 17; ewes came into estrus between Day 17 and Day 28. Table 4 shows the frequency of ewes in estrus and the mean time from introduction of rams to estrus onset. A greater percentage of ewes from Groups D5, D3, D1, and C3 than from Group D0 came into estrus during the observation period ( $P < 0.001$ ). Most ewes from Groups D5, D3, and D1 came into estrus earlier (Days 17 to 20; Table 4). Conception rates (pregnant ewes/ewes in estrus) were 75.0% (27/36), 90.9% (10/11), 84.8% (28/33), 77.4% (24/31), and 81.3% (26/32), respectively, for Groups C3, D0, D1, D3 and D5, respectively ( $P > 0.05$ ). Pregnancy rates (pregnant ewes/treated ewes) were 71.1% (27/38), 27.0% (10/37), 73.7% (28/38), 63.2% (24/38), and 65.0% (26/40) for the same groups (D0 versus C3, D1, D3, and D5,  $P < 0.01$ ).

## 4. Discussion

Our results show that short-term MAP priming is as effective as traditional long-term priming. Moreover, sponges containing a lower dosage of MAP than contained in commercial sponges can be used in 6-day priming. The use of MAP priming allowed us to advance estrus by 2 weeks in more than half of primed ewes (Experiments 1 and 2), with a conception rate ~60% (Experiment 1). In the remaining primed ewes, in accordance with previous observations after priming with fluorogestone and progesterone [16], estrus was delayed to the first half of the second estrous period (Days 17 to 20), probably as a consequence of a first ovulation on Days 1 to 3, followed by an NLP. Results obtained with

short-term priming, previously also observed in combination with eCG [6,12], constitute an important advantage when working under field conditions. The possibility of using vaginal sponges with a lower MAP content than that of commercial sponges without affecting fertility in sheep has previously been reported in other studies on cyclic ewes [9,17] and anestrus ewes stimulated with eCG (long-term priming: 10, 11; short-term priming: 12).

In the three experiments, we observed that ewes in the control groups exhibited delayed estrus with an unusual concentration during the expected period for the second estrous peak (Days 21 to 25; mean time from introduction of the rams to onset of estrus:  $24.3 \pm 0.4$  days,  $22.0 \pm 0.3$  days, and  $20.9 \pm 0.4$  days for Experiments 1, 2, and 3 respectively; Fig. 1). An explanation for this finding could be found in the observed progesterone patterns (Experiment 2): most control ewes responded to RE with an SLP followed by an NLP or a delayed NLP; however, very few entered an NLP immediately after the introduction of the rams. The phenomenon of unprimed ewes which do not ovulate immediately after ram introduction and in whom ovulation is delayed by 5 to 9 days has been previously reported by Hunter and Lishman [18] and Fulkerson et al. [19] and confirmed in an ultrasonographic study by Ungerfeld et al. [20]. As progestogens promote follicular turnover [21,22], more primed ewes should present a growing follicle when rams are introduced. In other words, a greater number of ewes should ovulate immediately instead of delaying ovulation until the next follicular wave offers a growing follicle. Some of the other responses to ram introduction previously reported, such as an SLP or two SLPs not followed by an NLP, may be related to some degree of luteinization of follicles [20].

A single MAP dose can substitute for progesterone administration at the time that rams are introduced. As a matter of fact, if MAP is given in adequate dosages and at an appropriate time (2.5 mg administered 3 or 5 days before the introduction of the rams), it ensures that estrous behavior will be concentrated on Days 17 to 19, meaning that the first ovulation is followed by an NLP. In the present study, ewes treated on Day -1 showed a more extended response, and some ewes treated when rams were introduced (Group D0) did not come into estrus until Day 28. This suggests that MAP continues to act for more than 1 day, thus inhibiting or delaying the pituitary response to the introduction of the rams.

In general, MAP administration, either by sponges or as a single treatment, prevents the occurrence of SLPs. There are at least two ways in which MAP can modify ovarian activity. Firstly, progestogens promote follicular turnover [21,22], which means that a young healthy follicle may be present in primed ewes when rams are introduced. Secondly, there is also experimental support for a direct effect of progesterone (and presumably MAP) at the ovarian level, ensuring the development of an NLP in anestrus ewes stimulated to ovulate with gonadotropin-releasing hormone (GnRH) [23].

In the present study, the duration of the NLPs was related to their onset (Experiment 2). Luteal phases that began late were shorter, and luteolysis coincided with the beginning of the second estrous period. The stimulus of the mating activity may have triggered an increase in LH pulsatility and thus an increase of estrogen secretion and start of luteolysis. Chemineau [24] reports that luteolysis is advanced when a buck is introduced to cyclic goats during the late luteal phase. A similar shortening of the luteal phase is observed after male stimulus in mice [25] or pheromone stimulus in humans [26].

We conclude that progestogen priming allows a significant percentage of ewes to display estrus during the first day after the introduction of the rams, and prevents the formation of

SLPs; that short-term MAP priming may be used with results similar to those achieved by traditional long-term priming; and that as little as 20 mg of MAP in vaginal sponges is enough to ensure the same reproductive response than when 60 mg of MAP are used. Additionally, after a single administration of MAP 3 to 5 days before the introduction of rams, estrus is displayed synchronized on Days 17 to 19.

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