

## Effect of estrus synchronization and artificial insemination on reproductive performance of Merino sheep

## Efeitos da sincronização do estro e da inseminação artificial sobre a performance reprodutiva de ovelhas Merino

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### SUMMARY

The experiment was conducted to determine the reproductive performance (lambing rate and litter size) of sheep artificially inseminated at a synchronized estrus in relation to rams, dose of PMSG and number of artificial inseminations performed. During spring cyclic Merino adult ewes were treated with intravaginal sponges impregnated with 60 mg medroxyprogesterone acetate (MAP). After 14 days sponges were removed and the females received an intramuscular injection of either 375 or 400 IU pregnant mare serum gonadotropin (PMSG). Estrus detection was performed with vasectomized rams. Females in estrus were cervically inseminated with fresh diluted semen from different rams either once or twice. Data on lambing rate and litter size were recorded. Lambing rate was not affected by rams or number of inseminations. A higher proportion of ewes that received 400 IU PMSG lambed compared with those receiving 375 IU PMSG (76.47% v 54.32%;  $p < 0.01$ ). Litter size was not affected by any of the variables under study.

**KEY-WORDS:** Sheep. Reproductive performance. Artificial insemination. PMSG. Rams.

### INTRODUCTION

Synchronization of estrus in sheep has been achieved by the use of intravaginal sponges containing synthetic progestagens such as medroxyprogesterone acetate (MAP; 6 $\alpha$ -methyl-17 $\alpha$ -acetoxy-pregne-4-ene-3,20-dione)<sup>1,2,7,13</sup>.

Several synchronization treatments incorporate an injection of a low dose of pregnant mare serum gonadotropin (PMSG) at the end of the progestagen treatment in order to obtain a more precise and reliable synchronization of estrus since it increases the occurrence of ovulation and improves the post-treatment fertility in small ruminants<sup>5,10</sup>. Therefore rate of ovulation is affected by the PMSG dose level employed<sup>4,10,15</sup>. An adequate dose of PMSG improves prolificacy, but the use of a high dose produces multiple gestations and thus, an increase in fetal or lamb mortality<sup>3</sup>. So, to avoid non-desirable fetal losses and large litter sizes, the dose level of such gonadotropin has to be precisely adjusted according to the season of the year and the physiological state of the ewes<sup>5</sup>.

Rams are another factor that can influence the reproductive performance of a flock. In this way Crempien et al.<sup>6</sup> found differences in fertility of ewes naturally mated due to rams.

Fertility can be also improved with the practice of a

double artificial insemination (AI) in the same estrus. The effect of this double AI depends upon the time when the first AI is performed in relation to ovulation. So, the effect of the double AI is more reliable when the first AI is realized at the beginning of estrus than when it takes place in the middle or at the end of estrus. Therefore double AI prevents from the possibility that insemination is practiced too early in relation with ovulation<sup>8</sup>.

The objectives of this study were to determine the reproductive performance (lambing rate and litter size) of adult ewes artificially inseminated at a synchronized estrus in relation to rams, dose of PMSG and number of artificial inseminations performed.

### MATERIAL AND METHOD

#### Animals and management

Cyclic Merino adult ewes were used in this study. The experiment was conducted during spring. The animals were managed under the same conditions on one farm. They were kept under natural field conditions, having access to good quality grasses and maintained in good health.

#### Synchronization method

Females were treated with 60 mg MAP in impregnated polyurethane sponges. Polyurethane sponges were prepared

by the method already reported by Robinson<sup>14</sup>.

Pessaries were inserted deep into the vagina and left in place for 14 days.

At sponge withdrawal, ewes received an intramuscular injection of either 375 or 400 IU PMSG.

### Estrus detection

The onset of estrus was carried out by the use of vasectomized rams in a ratio of 5%. Rams were painted with a vegetable dye mixed in a vaseline base so that ewes that were mounted could be identified. The males were introduced in the herd after pessary removal and for a total period of 96 hours.

Ewes were inspected for the presence of marks at 4-hours intervals.

### Artificial insemination

Merino rams (n=3; identified as A, B and C) with probed fertility were utilized in the artificial insemination program.

Semen from each ram was collected using an artificial vagina. Having determined seminal characteristics, it was diluted with egg yolk-tris-fructose extender.

Ewes that had exhibited estrus (n=166) were allocated at random to one of the three rams for artificial insemination and they were inseminated with fresh diluted semen containing a dose of 300 millions total sperm. Cervical artificial insemination was performed either once (7 to 11 hours after estrus detection) or twice (3 to 4 hours after estrus detection and 8 to 9 hours after first insemination).

### Reproductive performance

Percentage of ewes lambing to artificial insemination at the synchronized estrus (lambing rate) and number of lambs born per female lambing (litter size) were used as measures of reproductive performance.

Lambing was determined by daily observation and identification of ewes in small lambing plots.

### Statistical analysis

The GENMOD procedure of SAS was used to analyze the effects of rams, dose of PMSG and number of artificial inseminations on lambing rate and litter size.

For lambing rate data a binomial distribution with a logit link function was set. For litter size data a Poisson distribution with a log link function was set. Deviance criterion was employed for assessing goodness of fit in each case.

A model that included all main effects and interactions was tested. As interactions were found to be non-significant, they were excluded from the model. So a model with all main effects was finally used.

## RESULTS AND DISCUSSION

Data on reproductive performance of adult ewes in relation to PMSG dose level are given in Tab. 1.

A previous study performed by Smith, Boland and Gordon<sup>15</sup> reported 66,3% lambing rate after cervical AI following a progestagen (MAP)-PMSG (375 IU) treatment in lowland ewes. In the present study, working with other breed (Merino) a 54.32% lambing percentage was obtained when ewes were treated with the same gonadotropin dose.

Maxwell<sup>11</sup> and Maxwell and Hewitt<sup>12</sup> carried out studies on AI of mature Merino ewes during spring. In their experiments females were synchronized with sponges impregnated with fluorogestone acetate (FGA) and 400 IU PMSG were given at the end of treatment. After cervical AI with fresh diluted semen, they achieved a 47,5% lambing and 60% pregnancy rate, respectively. However, in our study a higher lambing percentage (76,47%) was obtained when 400 IU PMSG were injected.

When analyzing data obtained in the present study, a significantly higher proportion of adult ewes that received 400 IU PMSG lambed compared with those receiving 375 IU PMSG (76.47% v 54.32%; p<0.01).

**Table 1**

Reproductive performance of adult ewes in relation to PMSG dose level.

	375 IU	400 IU
No. inseminated	81	85
No. lambing (%)	44 (54.32) <sup>a</sup>	65 (76.47) <sup>b</sup>
Litter size	1.36 <sup>c</sup>	1.45 <sup>a</sup>

Values with different letters within each file differ significantly (p<0.01).

Same letters within a file do not differ significantly (p>0.05).

**Tabela 2**

Reproductive performance of adult ewes in relation to the number of artificial inseminations performed.

	Single AI	Double AI
No. Inseminated	104	62
No. lambing (%)	67 (64.42) <sup>a</sup>	42 (67.74) <sup>a</sup>
Litter size	1.46 <sup>a</sup>	1.33 <sup>a</sup>

Same letters within a file do not differ significantly (p>0.05).

**Tabela 3**

Reproductive performance of adult ewes in relation to rams used for artificial insemination.

	A	B	C
No. Inseminated	54	53	59
No. lambing (%)	36 (66.67) <sup>a</sup>	32 (60.38) <sup>a</sup>	41 (69.49) <sup>a</sup>
Litter size	1.44 <sup>a</sup>	1.38 <sup>a</sup>	

Same letters within a file do not differ significantly (p>0.05).

Smith, Boland and Gordon<sup>15</sup> informed 1,67 lambs per ewe lambing when a dose of 375 IU was tested in lowland breeds, whereas litter size obtained in our study was 1,36. This difference in reproductive response could be attributed to breeds. Maxwell<sup>11</sup> reported a litter size of 1,46 in Merino ewes treated with 400 IU PMSG, which was similar to 1,45 lambs born per ewe lambing in the present experiment for the same PMSG dose. When comparing data obtained in our study, the PMSG dose level employed did not significantly influence ( $p>0.05$ ) the number of lambs born per female lambing.

Among ewe breeds, Merino is considered to be a low prolific one<sup>8</sup>. The use of PMSG increases ovulation rate and thus the incidence of multiple births and the chances of pregnancy toxemia. In Argentina, Merino ewes breeding is mainly carried out under extensive conditions, so it is necessary to avoid these problems through an adequate adjustment of the gonadotropin dose.

Although the addition of PMSG to an estrus synchronization program tends to elevate costs, our data show a clear advantage of increasing a dose as low as 25 IU (from 375 to 400 IU) of such gonadotropin upon lambing rate without the risk of augmenting too much the number of lambs born per female lambing. However, an attempt must be made in terms to consider that a great variation on the ovulatory response could exist when PMSG is used for

estrus synchronization in response not only to differences among breeds and individuals, but also to batches of PMSG.

Tab. 2 shows the effect of the number of inseminations performed on reproductive performance of adult ewes.

The number of artificial inseminations (one or two) did not significantly affect the lambing rate or the litter size ( $p>0.05$ ). However, an improvement of 6-10% in lambing rate was previously reported<sup>8</sup> following a double insemination.

In view of these data it could be suggested that under an intensive regime of estrus detection there are no advantages for a double artificial insemination. So, a single AI could be practiced to obtain similar results without increasing labor. Moreover, rams could be used in a less intensive way for semen collection or a lower proportion of them could be required.

The effect of rams on reproductive performance is shown in Tab. 3.

Crempien, Barría and Gordon<sup>6</sup> found differences in fertility of ewes naturally mated in relation to rams. Conversely, in the present study neither lambing rate nor litter size differed among the three rams used for insemination ( $p>0.05$ ). The reason for this may be that males to be used for AI are previously tested (libido and semen characteristics) and this may not occur when natural service is to be performed.

## RESUMO

Foi estudada a resposta reprodutiva (taxa de parição e tamanho de leitigada) das ovelhas inseminadas artificialmente após o tratamento de sincronização do estro em relação à carneiros, dose de PMSG e número de inseminações artificiais. Ovelhas adultas Merino em estação reprodutiva (primavera), foram tratadas com esponjas intravaginais impregnadas com 60 mg acetato de medroxiprogesterona (MAP) para sincronização do estro. As esponjas foram retiradas após 14 dias e foram administradas i.m. 375 ou 400 UI gonadotrofina da égua prenhe (PMSG). Utilizaram-se carneiros vasectomizados para a detecção do cio. As ovelhas que apresentaram cio foram inseminadas artificialmente com sêmen fresco diluído de carneiros, uma ou duas vezes. A taxa de parição não foi afectada pelos carneiros ou pelo número de inseminações. A taxa de parição foi maior nas ovelhas tratadas com 400 UI PMSG que aquelas tratadas com 375 UI (76.47% v 54.32%;  $p<0.01$ ). O tamanho de leitigada não sofreu influência das variáveis estudadas.

**PALAVRAS-CHAVE:** Ovelhas. Resposta reprodutiva. Inseminação artificial. PMSG. Carneiros

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