

Residual levels on medroxyprogesterone acetate-impregnated sponges after estrus synchronization treatment and their relationship with fertility in cyclic goats

Níveis residuais em esponjas impregnadas com acetato de medroxiprogesterona, após tratamento de sincronização do estro e sua relação com a fertilidade nas cabras em período reprodutivo

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SUMMARY

A pool polyurethane sponges impregnated with medroxyprogesterone acetate (MAP) was prepared. Real level of progestagen on sponges was checked prior to sponges insertion. 13 cyclic goats were intravaginally treated with MAP-impregnated pessaries for the synchronization of estrus. After 14 days treatment, sponges were removed. 48 hours post-withdrawal, goats which exhibited estrus were artificially inseminated. Residual levels of MAP on removed sponges were measured by spectrophotometry at 241 nM. and examined in relation to their fertility. The real dose of MAP was in average $62 \text{ mg} \pm 2 \text{ mg}$. High levels ($x = 32.46 \text{ mg} \pm 9.84 \text{ mg}$) of residual MAP were found on sponges following treatment. The percentage of estrus synchronization was 92.31% and the pregnancy rate was 69.23%. Pregnant goats had significantly higher ($p \leq 0.01$) residual amounts of hormone ($x = 35.56 \text{ mg} \pm 6.06 \text{ mg}$) remaining on sponges than non-pregnant goats ($x = 20.00 \text{ mg} \pm 10.58 \text{ mg}$). A high positive correlation ($r = +0.7158$) was found between residual MAP and fertility. It was concluded that there is a relation between residual level of MAP and the fertility of the synchronized estrus in cyclic goats.

UNITERMS: Goats; Estrus synchronization; Medroxyprogesterone acetate; Sponges; Fertility.

INTRODUCTION

Synchronization of estrus in sheep and goats has been accomplished by means of intravaginal sponges impregnated with natural progesterone^{16,21}, as well as with synthetic progesterone such as fluorogestone acetate (FGA; 9a-fluoro-11 β -hydroxy-17a-acetoxy-pregne-4-ene-3,20 dione)^{1,2,4,5,8,10,13,19,21,23,24,26} or medroxyprogesterone acetate (MAP; 6a-methyl-17a-acetoxy-pregne-4-ene-3,20 dione)^{1,3,4,6,7,9,19,20,24,25}.

Degree of success in synchronizing estrus varies among studies but the treatment is generally very effective^{5,12,14,17,18}. However, reduced fertility at synchronized estrus has been reported^{1,5,11,15,21}. The aim of this study was to determine the residual levels on MAP-impregnated sponges after estrus synchronization treatment and their relationship with fertility in cyclic goats.

MATERIAL AND METHODS

Animals and management

Sixteen, cyclic multiparous goats of mixed breeds (Saanen, Nubian and Criolla) ranging from 2 to 4 years of age were used in this study. The animals were kept in natural field conditions, access to good quality mixture pasture and maintained in good health.

They were managed in one farm, under the same conditions. These females were randomly assigned in two groups:

Group I (control; $n = 3$): goats were treated with polyurethane vaginal sponges without MAP.

Group II (treated; $n = 13$): goats were treated with polyurethane vaginal sponges impregnated with 60 mg MAP approximately.

Pessaries preparation and synchronization method

Polyurethane sponges were prepared by the method already reported by Robison²¹ (1965).

A total of 32 sponges were divided into four groups:

Group A ($n = 13$): sponges impregnated with MAP dissolved in 3 ml of ethylic alcohol 96%, used to check the real dose of MAP on sponges prior to sponge treatment insertion.

Group B ($n = 13$): sponges impregnated with MAP dissolved in 3 ml of ethylic alcohol 96%, inserted deep into the vagina of each doe of group II and left in place for 14 days.

Group C ($n = 3$): sponges impregnated with 3 ml of ethylic alcohol 96% without MAP, used as control of MAP dose of group A.

Group D ($n = 3$): sponges impregnated with 3 ml of ethylic alcohol 96% without MAP, inserted intravaginally in goats of group I and left in place for 14 days.

The ethylic alcohol 96% used during this study came from a pool of 10 lts.

Quantification of real and residual MAP

At withdrawal, each pessary from groups B and D was placed into a sterile flask until assay in order to determine the residue remaining MAP on removed sponges used for estrus synchronization. Real and residual levels of MAP were measured by spectrophotometry. Absorbance values for known concentrations of MAP (0 to 0.0035%) were determined at the wavelength of 241 nm. to construct a standard curve.

Sponges were washed with 100 ml of ethylic alcohol 96% to extract the MAP contained. Each solution was filtered by using SS blue ribbon filter paper. 0.5 ml of the filtered solution was taken and completed up to 10 ml with alcohol 96%. Final volume was divided in order to obtain duplicates of each sample.

Absorbance value of each duplicate was measured by spectrophotometry, and its MAP concentration was obtained from the standard curve.

Real and residual levels of the synthetic progestagen were determined as follows:

$$a = b \times d$$

where a = mg MAP.

b = MAP sample concentration (measured by spectrophotometry).

d = 1 : 20 dilution factor.

Estrus detection

Does were examined for estrus within 48 hours following sponge withdrawal. Detection of estrus was carried out by looking at their behavior and estrus signs (vulval swelling, mucus discharge, vaginal erythema and wagging tails and by the use of a buck fitted with an apron to prevent intromission.

Artificial insemination

Semen from a buck with probed fertility was collected by using an artificial vagina.

Having determined seminal characteristics, it was diluted with egg yolk-tris-fructose extender.

48 hours post-treatment, goats from group B that had exhibited estrus, were artificially inseminated with fresh, diluted semen. A dose of 0.25 ml containing 400 million total sperm was deposited deep into the vagina.

Pregnancy detection

Goats with non-return to estrus following 21 days artificial insemination were assumed to be pregnant. Sixty days after artificial insemination pregnancy was confirmed by ultrasound.

Evaluation of results

Statistical differences for residual MAP, in respect to fertility of the synchronized estrus were determined by Student's t-test.

Analysis of correlation (Pearson) was done between residual MAP and fertility of the synchronized estrus.

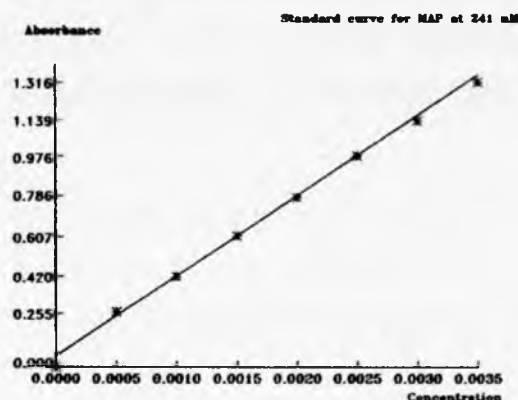


Figure 1

Illustrates the absorbance values of the standard curve for known concentrations of MAP as determined by spectrophotometry at 241 nm.

RESULTS

Tab. 1 shows absorbance, concentration, residual and uptake levels of MAP, estrus synchronization and fertility of goats from group II.

Tab. 2 summarizes estrus synchronization rate, pregnancy rate and residual and uptake MAP levels in relation to the reproductive performance.

A high positive correlation ($r = +0.7158$) was found between residual MAP and the fertility of the synchronized estrus.

DISCUSSION

The mean real MAP level found on sponges from Group A was $62 \text{ mg} \pm 2 \text{ mg}$.

The mean residual MAP level found on removed sponges from all treated goats following treatment during 14 days was $32.46 \text{ mg} \pm 9.84 \text{ mg}$, being the uptake level $29.54 \text{ mg} \pm 9.84 \text{ mg}$ (Table 2). The 47.65% of the MAP dose remained on pessaries following 14 days treatment. Mac Donnell¹⁶ (1985) detected a 36% of the 500 mg progesterone dose used in sponges to synchronize estrus in ewes, after 17 days treatment.

Percentage of estrus exhibition within 48 after hormonal treatment was 92.31% (Tab. 2). The high degree of synchrony achieved with this synthetic progestagen has been reported by other authors in goats^{3,7,17,18} and ewes^{1,19}.

Mean residual MAP level for goats that exhibited estrus was $31.67 \text{ mg} \pm 9.83 \text{ mg}$, being the uptake level $30.33 \text{ mg} \pm 9.83 \text{ mg}$ (Tab. 2). The goat that was not in heat following sponge withdrawal had a residual value of 42 mg MAP, having absorbed 20 mg MAP during treatment (Tab. 2).

Conception rate based on non-return to estrus and ultrasound-proof was 69.23% (Tab. 2), similar to that reported by Alaçam *et al.*³ (1985) goats.

Pregnant goats had significantly higher ($P \leq 0.01$) residual amounts of MAP ($x = 35.56 \text{ mg} \pm 6.06 \text{ mg}$) remaining on pessaries than non-pregnant goats ($x = 20.00 \text{ mg} \pm 10.58 \text{ mg}$).

Table 1

Residual and uptake MAP, estrus synchronization and fertility of 13 cyclic goats treated with MAP-impregnated intravaginal sponges for 14 days. Buenos Aires, Argentina, 1995

nº	Absorbance	Concentration	Residual (%)	Uptake	Estrus MAP(mg)	Fertility MAP(mg)
1	0,553	0,0014	28	34	+	+
2	0,718	0,0018	36	26	+	+
3	0,177	0,0004	8	54	+	-
4	0,820	0,0021	42	20	-	-
5	0,801	0,0020	40	22	+	+
6	0,570	0,0014	28	34	+	-
7	0,853	0,0022	44	18	+	+
8	0,710	0,0018	36	26	+	+
9	0,644	0,0016	32	30	+	+
10	0,529	0,0013	26	36	+	+
11	0,692	0,0018	36	26	+	+
12	0,813	0,0021	42	20	+	+
13	0,482	0,0012	24	38	+	-

Table 2

Amount of hormone (MAP) remaining and absorbed, estrus response and fertility of 13 cyclic goats treated with MAP-impregnated intravaginal sponges for 14 days. Buenos Aires, Argentina, 1995

Reproductive performance	Nº goats	Residual MAP (mg)	Uptake MAP (mg)	% Estrus synchronization	% Pregnancy rate
Total Nº	13	32,46±9,84	29,54±9,84		
Mated	12	31,67±9,83	30,33±9,83	92,31	
Not mated	1	42,00±0,00	20,00±0,00	7,29	
Pregnant	9	35,56±6,06(a)	26,44±6,06(a)		69,23
Not pregnant	3	20,00±10,58(b)	42,00±10,58(b)		30,77

a, b = Different letters of a same column have a significant ($P \leq 0,01$) difference for residual and uptake levels of MAP in relation to pregnancy.

CONCLUSION

In the comprehensive study of synthetic progesterone derivatives impregnated in sponges, it was suggested²² that the progestagen dose required to inhibit ovulation during treatment is lower than that required to synchronize estrus, and that the dose required for such conditioning is still lower than the one to obtain a fertile response of ewes to such treatment, for estrus synchronization.

1. In this study it was possible to determine the existence of a residue of MAP on sponges removed from goats vaginas after 14 days of permanence. Therefore, the 60 mg dose of such synthetic progestagen conventionally used for estrus synchronization in small ruminants, by means of intravaginal pessaries, is higher than that uptake by them.
2. There is a relation between residual levels of MAP and fertility of the synchronized estrus in goats. A higher residual level of MAP was detected remaining on pessaries from pregnant goats than on pessaries from non-pregnant ones.
3. There are individual differences in the amount of MAP uptaken.

RESUMO

Preparou-se um conjunto de esponjas de poliuretano embebidas com acetato de medroxiprogesterona (MAP). O nível real do progestágeno nas esponjas foi checado com anterioridade à inserção das esponjas do tratamento. Treze cabras em período reprodutivo foram tratadas com esponjas intravaginais embebidas em MAP para sincronização do estro. As esponjas foram retiradas após 14 dias de tratamento. As cabras que apresentaram cio foram inseminadas artificialmente 48 horas depois de retiradas as esponjas. Os níveis residuais de MAP nas esponjas retiradas foram medidos por espectrofotometria a 241 nM e examinados em relação à fertilidade. A dose real de MAP teve como média $62 \text{ mg} \pm 2 \text{ mg}$. Níveis elevados ($x = 32,46 \text{ mg} \pm 9,84 \text{ mg}$) de MAP residual foram encontrados em esponjas após o tratamento. A porcentagem de sincronização dos cios foi 92,31% e a taxa de prenhez, 69,23%. As cabras emprenhadas tiveram um nível residual do hormônio nas esponjas significativamente maior ($P \leq 0,01$) ($x = 35,56 \text{ mg} \pm 6,06$) que as cabras não emprenhadas ($x = 20,00 \text{ mg} \pm 10,58 \text{ mg}$). Encontrou-se uma alta correlação ($r = +0,7158$) entre MAP residual e fertilidade. Conclui-se que há uma relação entre o nível residual de MAP e a fertilidade dos cios sincronizados nas cabras em período reprodutivo.

UNITERMOS: Cabras; Sincronização do estro; Acetato de Medroxiprogesterona; Esponjas; Fertilidade.

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