# EFFECTS OF A PMSG TREATMENT ON BREEDING RESULTS OF ARTIFICIALLY INSEMINATED RABBITS

Bourdillon A.<sup>1</sup>, Chmitelin F.<sup>1</sup>, Jarrin D.<sup>1</sup>, Parez V.<sup>2</sup>, Rouillère H.<sup>1</sup>

SANDERS ALIMENTS - 17, quai de l'Industrie - 91200 ATHIS-MONS - FRANCE
 INTERVET S.A. - 43, Avenue Joxé - 49100 ANGERS - FRANCE

# ABSTRACT

The effect of a 30 UI intra-muscular injection of PMSG 48 hours before artificial insemination (AI) was compared with a control injection on female rabbits who were inseminated 10 days after parturition. We noticed that the fertility rate increased by 8 points (p < 0.05) (76.3 % vs 68.3 %) among the nursing females injected with PMSG, while this very same treatment didn't seem to be efficient on dried females (79.1 % vs 85.7 % - not statistically significant). However, PMSG prompts an increase in the prolificity in terms of number of rabbits born by litter, whatever the physiological stage of the female.

We analyzed the effects of a PMSG treatment, depending on the number of litters before AI. We noticed a very large impact of the treatment among the females who were in their first lactation (a fertility rate of 57,6 % vs 29,4 % and 13,29 vs 10,56 rabbits born by litter). But the effect of the treatment is not relevant on multiparous females (a fertility rate of 79,6 vs 76,6 % and 11,03 rabbits born by litter vs 10,35). We may emphasize that the circulating antibodies rate tends to increase after 3 PMSG injections in a row, although this increase is not statistically relevant. But, after the fourth injection, the injected females fertility doesn't fall below the average fertility of the control batch.

#### INTRODUCTION

Unlike most domestic species, female rabbits don't have a classical oestrus cycle with regular heats during which ovulation would occur spontaneously. One has been reckoning for a very long time that does are in an ever-lasting oestrus stage and that ovulation is prompted by mating (HAMMOND & MARSHALL, 1925). But, about behaviour, HILL and WHITE (1933-1934) and MORET (1980) showed that oestrus stage, where does accept mating, and dioestrus stage, where they refuse it, occur alternatively.

However, the length and periodicity of these mating cycles seem to vary very much, depending on the does. And the hormonal factors that run it aren't well known. The oestrus stage would be triggered by the coming out at the surface of the ovary of matured or pre-ovulatory follicles. THEAU-CLEMENT *et al.* (1990) showed indeed that ovulation frequency and fertility are very high among receptive does and are significantly lower among non receptive ones.

In rabbit breeding, fertility is scanned thanks to the perception, through abdominal palpation, of embryonic vesicles which takes place 10 to 14 days after mating. In natural mating, females who aren't in an oestrus stage are presented to males every day until mating occurs. Therefore, breeding records are calculated only on the receptive females. In AI, on the other hand, all females are inseminated, be they receptive or not, and results are calculated on the overall number of females which may explain why results fall when a rabbit breeder switchs from natural mating to AI. A synchronization of oestrus that would enable an insemination of females who would all be receptive would therefore be very useful.

So as to try to trigger receptivity of females, 48 hours before AI we gave a PMSG treatment (gonadotropin extracted from the serum of pregnant mare). This glycoprotein, which has a large molecular weight, stimulates ovarian interstitial cells growth as long as development and maturing of follicles (FSH effect). However, its weak LH effect also put pressure on the ovulation rate.

# MATERIALS AND METHODS

The experiment was led over a 6 month reproduction period in Sourches (France), the research facility of Sanders Company. The first mating took place in May 1991 and the last weanings were achieved in October 1991.

#### Treatments

One hundred does were split into two groups of 50 females according to the number of previous pregnancies. One group received an intra-muscular (i.m.) injection of 35 UI of PMSG (Chrono-gest PMSG 600® - Intervet) in the leg 48 hours before AI, while the control group received on the same day an i.m. injection of the physiological solvent used to prepare the PMSG. So as to study the impact of repeated PMSG injection on breeding records, the trial was conducted with 4 AI in a row.

### Animals

French hybrid does (Hyplus 0) were fed ad libitum with a diet containing 18 % of crude protein and 10.8 MJ DE/kg from parturition and during 18 days after parturition and with a diet containing 15,5 % crude protein and 10,0 MJ DE/kg from 18 days of lactation to the next parturition. The does were first inseminated when 16 weeks old. The nulliparous females who are generally good at being artificially inseminated were put off. One hundred does out of our experimental 270 females farm stock were used (n = 50 per treatment) in the trial. The does who died or were culled during the test were superseded by new females who therefore intered the test protocol, after the second AI. Does which were not pregnant 3 times in a row were eliminated. Females who showed signs of diseases or underproductivity (less than 5 weaned rabbits per litter 2 times in a row) consecutively were also culled. Litter size was standardized to 10 tabbits at birth.

# Housing

Does were housed in windowless buildings of 100 places on flat-deck cages for parturition (77  $\times$  48 cm) and 16 h of light per day was provided. A minimum temperature of 16°C was maintained. A dynamic ventilation system with low extraction was used. Manure was collected in a deep pit and removed at the end of each fattening period.

The 100 females were inseminated on the same day : it's the reason why insemination as well as parturition and weaning took place on the same day for the 100 does. At each weaning, the young rabbits stayed in the room in the flatdeck cages for fattening ( $88 \times 48$  cm) and the does went into another compartment that had been in a sanitory vacuum for 10 to 15 days, where they gave birth 12 to 15 days later. On day 28 of gestation does received their metal nest box. They were set outside the cage and wood shavings were displayed. The males were housed in a separate compartment and received only 8 h of light.

# Breeding shedule

All the does were inseminated 10 days after parturition. Palpation was performed at day 14. Non-pregnant does were reinseminated 21 days after parturition and therefore received 2 injections of PMSG at an interval of 3 weeks instead of 6 weeks for pregnant females.

The does were artificially inseminated with the mixed sperms of 10 bucks (polyspermy) with a dilution rate of 4 in Dilap 2000 (IMV). The insemination took place 2 to 3 hours after collection of the fresh sperms. Just after insemination, the females were injected i.m. with 0,20 ml of GnRH (Fertagyl <sup>®</sup> - Intervet).

The following data were recorded on all does : fertility, birth rate, litter size total and number of born rabbits alive. We didn't test the number of weaned rabbits because the litters were standardized to 10 rabbits at birth. The culling and death rate wasn't taken into account either, because the variability of this king of data is so large that it needs to be studied on a very long period to be really relevant.

## Statistical analysis

Statistical analysis have been done on S.A.S. software 6.04 (SAS/STAT user's guide, 1988). Fertility results were analysed through the variance analysis for qualitative variables method. Birth results and the measures of the antibody rates were analysed through the GLM Process.

We analysed the impact of PMSG on breeding records (fertility, birth rate), taking into account the physiological stage of the females (nursing or not), the number of litter before AI and the number of PMSG injections.

#### RESULTS AND DISCUSSION

In overall 256 AI were performed in the control group and 255 in the PMSG group. The number of does dead or culled was identical in both groups (24 vs 25).

# Effect of PMSG according to physiological condition of the does at AI (Table 1)

For the analysis we separated the does in two groups, depending on the physiological condition at the AI. In one hand, females inseminated at 10 days postpartum are in lactation, on the other hand, females not pregnant at palpation 14 days post AI, reinseminated at 3 weeks post AI i.e. 3 days after weaning, are dried.

Fertility (number of positive palpations on number of inseminated females) improved (p<0,01)) among nursing females, thanks to PMSG. However, among dried females, fertility was very high in the control batch (85,7%) and PMSG therefore triggered no improvement. These results confirm the ones of THEAU-CLEMENT *et al.* (1991) which showed a higher fertility in AI, among the dried females than among the nursing ones. This could stem from a negative effect of the lactation as well as from a positive effect of a longer interval between two successive parturitions.

Prolificity increased by one rabbit by litter compared with the control batch, whatever the physiological status, nursing or not (p<0,05), which confirms the results of MAERTENS *et al.* (1983) in natural mating.

# Effect of PMSG according to the number of litter before AI. (Table 2)

We dispatched nursing females according to the number of litter before AI in order to analyse the possible interaction between treatment and birth rank. In our testing conditions, does who were inseminated during the lactation that follows the first parturition have lower fertility rate (30 %) than for multiparous females, which confirm the results of CHMITELIN *et al.* (1990). In that case, a PMSG injection 48 hours before AI doubles the fertility rate (p<0,05). Furthermore, we may underline that prolificity increased by 3 rabbits in this batch of does (p<0,05). Among multiparous females, on the other hand, there was no significant increase of fertility or prolificity (Table 3).

## Effect of PMSG according to the number of PMSG injections (Table 4)

We analysed the impact of repeated PMSG treatments on the evolution of rabbit fertility. This analysis was achieved on the group of multiparous and nursing females. Repetition of PMSG treatments, with less than 5 injections, doesn't fuel a fall of fertility below the average fertility of the control group (76,6 % - Table 3). That confirms the results of BONNANO *et al.* (1991).

The females who got 5 or 6 injections have a fertility rate of 70 %. However, the number of females (n=10) is not high enough to let us draw any relevant conclusions on the impact of PMSG beyond 4 injections. Prolificity is not weakened by repeated PMSG treatments (Table 4).

Furthermore, we took blood samples of 22 females who got 3 PMSG or solvant injections in a row, at a 42 days interval, so as to seek for the circulating anti PMSG antibodies rate (Table 5). We consider that there are some anti-PMSG antibodies when the OD (optical density), with a dilution of 1/640, is higher than 30 %.

We may notice that the PMSG batch is above this threshold and that the control batch is under it. However, the difference between the two batches is not relevant because two females of the control batch are above this threshold (50,6 and 75,4 %) while they never received any PMSG.

These results seem to confirm the conclusions of CANALI *et al.* (1991) who revealed an increase of the circulating antibodies rate after repeated injections of PMSG. However, the fertility results at the fourth injections (Table 4) are in contradiction with the CANALI results (CANALI *et al.*, 1991) which revealed a fall in the positive palpation rate at this stage.

# CONCLUSION

The use of an oestrus synchronization treatment with PMSG in a flock of rabbits inseminated 10 days after parturition, is efficient when treatment is achieved on lactating females. However, the treatment is useless on females who are not considered as pregnant after palpation and who are therefore inseminated after weaning. Furthermore, in our testing conditions, for nursing females, the effect of the treatment is highly relevant on the first lactation females, whose fertility records are very low in the control group. In that female batch, PMSG also enables an increase in prolificity of 3 rabbits by litter. However, the effect of treatment on multiparous females is not relevant.

Therefore, in our testing conditions, it seems that one may use PMSG only with primiparous does. However, our breeding records being at quite good a level (with a fertility rate of 76 % on average), it would be necessary to achieve an experiment on a flock in less good a condition, in order to confirm these results.

534

# Table 1: Effect of a PMSG injection on the breeding records of does accordingto their physiological status at AI

Physiological status	LACT	ATING	DRY		
Groups		Control	PMSG	Control	PMSG
Number of AI		200	212	56	43
Fertility	%	68,3a	76,3b	85,7b	79,1b
Total rabbits born/litter		10,37a	11 <b>,2</b> 9b	10,36a	11,23ab
Rabbits born alive/litter		9,49a	10,46b	9,66a	10,54b

a vs b : p<0,05 when on the same line

Table 2 : Effect of a PMSG injection on lactating does according to the numberof previous parturitions

Groups	Control	PMSG	Control	PMSG	Control	PMSG	Control	PMSG
Number of previous parturitions (n)	n=1		2≤ n ≤4		5≤ n <i>≤</i> 9		n ≥10	
Number of AI	34	33	54	51	58	56	54	72
Fertility %	29,4	57,6	69 <i>,</i> 8	75,5	82,8	83,3	79,6	81,9
Total rabbits born/litter	10,56a	13 <b>,29</b> b	10,91ab	11, <b>46</b> a	11,06ab	11,80ab	9,3a	10,33ab
Rabbits born alive/litter	9,80a	12,59b	10,15ab	10 <b>,5</b> 5ab	10,43ab	10,94b	8,14a	9,52a

a vs b vs c : P<0,05 when on the same line

Table 3: Effect of PMSG injection on lactating does primiparous or multiparous

		Primip	oarous	Multiparous		
Groups		Control PMSG		Control	PMSG	
Number of AI		38	34	166	179	
Fertility	%	29,4	57,6	76,6c	79,6c	
Total rabbits born/litter		10,56a	13,29b	10,35a	11,03a	
Rabbits born alive/litter		9,80a	12,59b	9,47a	10,19a	

a vs b vs c : P<0,05 when on the same line

Table 4 :Effect of repeated PMSG injections of multiparous nursing females<br/>on breeding records

Number of injections		1	2	3	4	5 et 6	Stat. Sign. <sup>(1)</sup>
Number of AI		43	43	50	33	10	
Fertility	%	79,0	90,7	76,1	81,3	70,0	NS
Total rabbits born/litter		10,68	10,11	11,08	11,79	11,14	NS
Rabbits born alive/litter		<b>9,2</b> 0	9,23	10,58	11,13	9,43	NS

(1) NS = not signif. different

# Table 5 : Circulating anti-PMSG antibodies after injections in a row

Groups		Control	PMSG	Stat. Sign. <sup>(1)</sup>
Number of females		11	11	
Optical Density at 1/640 dilution (	%)	22,27	37,64	NS

(1) NS = not signif. different

. A

536

#### REFERENCES

• BONNANO A., ALABISO M., ALICATA ML., 1991. Effetti del trattamento sincronizzante con PMSG su coniglie inseminate artificialmenti, Rivista di coniglicoltura N° 11, 29-32.

• CANALI C., BOTI C., ZAMPINI D., CASTELLINI C., BATTAGUNI M., 1991. Correlazione tra fertilita et titolo anticorpale anti PMSG di coniglie trattate ripetutamente con gonadotropine nel corso della lora carriera riproduttiva; Atti IX Congresso Nazionale ASPA, 671-678.

• CHMITELIN F., ROUILLERE H., BUREAU J., 1990. Performances de reproduction des femelles en insémination artificielle en post-partum, 5èmes journées de la recherche cunicole en France, Tome I, Comm. 4.

• HAMMOND J. et MARSHALL FHA., 1925. Reproduction in rabbit. Olivier and boyd. ed. London, 210p.

• HILL M., WHITE WE., 1933-1934. The growth and regression of follicules in the oestrus rabbit J. Physiology. London, 80, 174-178.

• MAERTENS L., OKERMAN F., GROOTE F. de, MOERMANS R., 1983. L'incidence de 2 méthodes de traitement hormonal sur le comportement sexuel et la fertilité de jeunes lapines. Revue de l'Agriculture, 1(36) 167-175.

• MORET B., 1980. Comportement d'oestrus chez la lapine. Cuniculture, 33, 159-161.

• SAS/STAT. User's guide, Release 6.04, 1988, SAS Institute INC, Cary, NC, USA, 190-282, 551-660.

• THEAU-CLEMENT M., BOLET G., ROUSTAN A., MERCIER P., 1990. Comparaison de différents modes d'induction de l'ovulation chez les lapines multipares en relation avec leur stade physiologique et la réceptivité au moment de la mise à la reproduction. 5ème journée de la recherche cunicole en France, Paris, Tome 1, comm. 6.

• THEAU-CLEMENT M., ROUSTAN A., 1991. Etude des relations entre réceptivité et lactation chez la lapine. Influence sur les performances de reproduction. Physiologie et pathologie du lapin de chair. Cycle AFTA, Paris 45-60.

537

