

## EFFECT OF MOTHER-LITTER SEPARATION ON REPRODUCTIVE PERFORMANCE OF LACTATING RABBIT FEMALES INSEMINATED ON DAY 4 OR 11 *POST PARTUM*.\*

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**ABSTRACT.**— Biostimulation of lactating rabbit females, through time limited controlled lactation, was compared to a conventional PMSG treatment. Experimental groups were composed with females separated from their litters for 0, 24, 36 or 48 hours before being inseminated on day 4 or 11 *post partum*. When biostimulation was applied, results were improved compared to non-stimulated females, especially fertility on day 4 *post partum*, after more than 24 hours of mother-litter separation ( $P < 0.01$ ). In either the 35 or 42 days reproductive rhythm, a single mother-litter separation for at least 36

hours, prior to AI, resulted in comparable fertility and prolificacy to those obtained in females treated with 20 IU of PMSG. Although litter survival was not affected, daily growth was slowed down and weaning weight reduced by 10% when litters were not allowed to suck for 36 or 48 hours ( $P < 0.01$ ). It is concluded that this technique could offer an alternative method to PMSG treatment in commercial units, in spite of the decrease in growth. However it should be advisable to evaluate at a long term its incidence on global productivity.

**RÉSUMÉ :** Effet d'une séparation ponctuelle entre la mère et sa portée, sur les performances de reproduction de lapines allaitantes inséminées 4 ou 11 jours après la mise bas.

L'intérêt d'une méthode de biostimulation de lapines allaitantes, par un contrôle ponctuel de la lactation, est comparé à un traitement classique de PMSG. Les groupes expérimentaux sont composés de lapines séparées de leur portée depuis 0, 24, 36 ou 48 heures, avant d'être inséminées le 4ème ou le 11ème jour après la mise bas. Quand la biostimulation est appliquée, les résultats sont améliorés par rapport aux lapines non stimulées, particulièrement la fertilité des lapines inséminées 4 jours *post partum* après plus de 24 heures de séparation ( $P < 0,01$ ). Quel que soit le rythme de reproduction, une séparation

ponctuelle des mères et de leur portée d'au moins 36 heures avant l'insémination conduit à des résultats de fertilité et de prolificité comparables à ceux obtenus par un lot de lapines traitées avec 20 U.I de PMSG. Bien que la viabilité des lapereaux ne soit pas influencée par la biostimulation, la croissance journalière diminue et le poids au sevrage est réduit de 10 %, quand l'allaitement est suspendu pendant 36 ou 48 heures ( $P < 0.01$ ). Malgré cette chute de croissance, les auteurs concluent que cette technique pourrait être une solution alternative au traitement PMSG dans les élevages commerciaux. Cependant, il conviendrait d'évaluer son incidence à long terme sur la productivité globale.

### INTRODUCTION

Rabbit artificial insemination (AI) is rapidly spreading along Europe and new problems linked to a massive utilisation are rising in rabbit farms.

Whereas nulliparous females can reach good results without PMSG (REBOLLAR *et al.*, 1994; REBOLLAR *et al.*, 1995a), to obtain good reproductive performance, stimulation of ovarian activity seems to be useful when lactating does are inseminated (REBOLLAR *et al.*, 1989). Usually lactating females are injected 20 IU of PMSG (ANGELI *et al.*, 1990; COLIN, 1992; ALVARIÑO and REBOLLAR, 1995) which is enough to improve the artificial insemination results. Nevertheless, several works alert about the reduced efficiency of PMSG when employed repeatedly and within short intervals of time (CANALI *et al.*, 1990; BOURDILLON *et al.*, 1992; REBOLLAR *et al.*, 1995b). Recently research has focused on alternative methods

of ovarian stimulation, as the administration of prostaglandins (FACCHIN *et al.*, 1992; ALVARIÑO *et al.*, 1995, PIMENTA *et al.*, 1996), or the so-called biostimulation techniques. Separation of mothers from their litters for short periods of time before AI has been found to increase fertility by 11-13%, although the litter weight at weaning could be negatively affected (PAVOIS *et al.*, 1994). At present different kinds of controlled lactation, closing nest boxes during 24 to 48 hours, are being tested in different countries and conditions. In this work this technique has been considered as a routine at the commercial level, studying if a short mother-litter separation from 24 to 48 hours, could replace a conventional PMSG treatment, considering the influence on AI results as well as on litter growth and survival.

### MATERIAL AND METHODS

A total of 2995 multiparous females of the Californian x NZW breed, housed in individual cages under controlled light/dark cycles (16h/8h) and fed *ad libitum* with a commercial pelleted diet, were used in

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**Table 1 : Reproductive performance after AI on day 4 *post partum***

Separation (h)	Mother-litter separation. : Experimental groups				PMSG Control
	0	24	36	48	0
Nb of AI	194	181	169	179	819
Fertility %	47.4 D	64.2 C	79.8 AB	81.8 A	74.9 B
Total Born	7.6 ± 0.4 C	7.9 ± 0.3 BC	7.9 ± 0.3 BC	8.3 ± 0.2 AB	8.5 ± 0.1 A
Born alive	7.1 ± 0.4 C	7.6 ± 0.3 BC	7.4 ± 0.3 BC	7.9 ± 0.3 AB	8.1 ± 0.1 A

A,B,C,D : P&lt;0.01

this study. The experimental design considered females inseminated on day 4 (experiment I) or 11 *post partum* (experiment II). In each case the control was a group of females inseminated following the usual procedure (ovarian stimulation by 20 IU of PMSG i.m., 48 hours before AI), and the experimental groups were composed with females not treated with PMSG separated from their litters by a metallic screen since 0, 24, 36 or 48 hours before insemination. When separation time was completed, kits were allowed to suck 5 minutes before the artificial insemination. In each experiment all females were inseminated the same day, starting at 8:00 p.m. and finishing at 12:00. Nest boxes were closed 24, 36 or 48 hours before, according to the expected insemination time. Only multiparous does, lactating 7–9 kits, were considered. Ejaculates were collected using artificial vagina and semen quality was assessed under microscope. Only ejaculates with over 60% of mobile sperm were selected. Sperm concentration was determined by a Burker's counting camera after pooling 5 selected ejaculates. By adding a commercial extender (MA 24, Laboratorios Ovejero, León Spain) a final sperm concentration of 25 million spermatozoa/ml was obtained. Does were inseminated with fresh semen. Ovulation was induced by 20 µg of GnRH (Inducel GnRH, Laboratorios Ovejero, León, Spain). One ml of diluted sperm was used to inseminate each female.

In experiment II, additionally, 263 litters were followed measuring weight and mortality, to evaluate

the possible negative effects on litter growth and survival. Litters were weighed on day 9 *post partum*, and this was considered as initial weight. Weaning weight was determined at the age of 32 days. Litter size was also recorded on day 9 and 32 *post partum*.

Statistics was performed using the CATMOD and GLM procedures of the SAS program, to compare fertility and prolificacy respectively. In the last case, means were compared using the Duncan test (SAS/STAT, 1985).

## RESULTS

Tables 1 and 2 show the effect of different duration of mother–litter separation on fertility and prolificacy when inseminations were performed on days 4 and 11, respectively. It seems clear that the absence of hormonal stimulation leads to a decrease in fertility more important on day 4 than on day 11 *post partum*. When biostimulation is applied, results improve especially when the separation mother–litter is over 36 hours. The increase in fertility is remarkable on day 4 *post partum*, improving after 24 hours of separation, and reaching similar (36h) or even higher values (48h) than obtained after PMSG treatment, (P<0.01). When AI was performed on day 11 *post partum*, the previous separation for 36 hours improves fertility compared to non–stimulated females (P<0.01), up to the level of PMSG group.

Litter size was only affected in females inseminated 4

**Table 2 : Reproductive performance after AI on day 11 *post partum***

	Mother-litter separation. : Experimental groups				PMSG Control
	0	24	36	48	0
Nb of AI	196	184	190	187	693
Fertility %	75.1 BC	78.6 B	85.6 A	81.6 AB	81.8 AB
Total Born	9.3 ± 0.3	8.8 ± 0.3	8.6 ± 0.2	8.9 ± 0.3	9.2 ± 0.1
Born Alive	8.9 ± 0.5	8.4 ± 0.3	8.2 ± 0.2	8.1 ± 0.2	8.7 ± 0.1

A,B,C: P &lt;0.01

**Table 3 : Effect of litter separation on young growth and survival until weaning (A.I on day 11 post partum)**

	Separation time (hours)			
	0	24	36	48
Nb. of litters	67	65	66	65
Initial weight (g)	111 ± 9	112 ± 16	107 ± 9	106 ± 9
Initial litter size	8.2 ± 0.1	8.0 ± 0.1	8.5 ± 0.1	8.1 ± 0.1
Weaning weight (g)	736 ± 12 A	700 ± 14 AB	663 ± 14 B	668 ± 12 B
Daily gain (gr.)	24 ± 0.4 A	23 ± 0.7 AB	21 ± 0.4 B	22 ± 0.4 B
Dead until weaning	0.94 ± 0.15	1.0 ± 0.17	0.84 ± 0.14	0.98 ± 0.15

A,B: p &lt; 0.01

days *post partum* (P<0.01), the worst results being obtained without any kind of stimulation. In the females inseminated after 48 hours of separation, the litter size (total born and born alive) was not different from that obtained after PMSG treatment.

The influence of mother-litter separation on growth and survival when AI was performed on day 11 *post partum*, is shown in table 3. Litter size and initial weight did not differ. Daily growth was slightly slowed down after separation of 36 hours or more (P<0.01), and the influence of mother-litter separation on weaning weight was significant (P<0.01). Compared to non-treated lactating females, a separation of 36 or 48 hours resulted in a reduction from 736 to 663 or 668 g. The fact that kits were separated for 24 to 48 hours, did not affect survival, so the mortality rate until weaning was not different from that of normal suckling kits.

## DISCUSSION

Data obtained in this work confirm previous results indicating that ovarian stimulation with PMSG improve performances when lactating females are inseminated (FACCHIN *et al.*, 1992 ; ALVARIÑO *et al.*, 1995).

The biostimulation through separation of mother and litters for short periods of time seems to be effective if the separation time exceeds 24 hours. In this case fertility increases to reach similar values to those obtained when PMSG is employed, confirming previous findings (PAVOIS *et al.*, 1994), although the effect is much more pronounced on day 4 than on day 11 *post partum*. Litter size is also affected on day 4, whereas it is not on day 11, indicating that the antagonism between reproduction and lactation is specially marked during the first 3-5 days of nursing. In fact, when suckling does not take place for 48 hours (days 2-4 of lactation) an increase of 0.7 total born

kits per litter was found, compared to normal lactating inseminated females.

Moreover, the separation does not seem to affect negatively the litter survival at least when it is performed around day 11 *post partum*. The influence on weaning weight is significant (P<0.01), as a decrease of 70 g (around 10% of normal weaning weight) was detected when litters were separated for 36 or 48 hours. In practice this should not be a problem, as part of this weight could be compensated during the fattening period. Unfortunately, no information was recorded in this work about growth and mortality when kits are separated from their mother at the age of 2-3 days. Authors personal observation indicates that this very young rabbits are able to stand a 48 hours separation without apparent modification in mortality rate.

In conclusion, whatever the studied rhythm of reproduction (35 or 42 days), a temporary mother-litter separation for at least 36 hours, previously to AI, offers similar results in fertility and prolificacy to that obtained in females treated with 20 IU of PMSG, although a decrease of 10% in weaning weight should be expected.

So, it seems that a limited controlled suckling could offer an alternative method to PMSG treatment in commercial units. This could be of interest in the next future, as natural methods of control of reproduction will be accepted better than the hormonal ones. Nevertheless, it should be advisable to confirm these results following a group of females along their reproductive life, using only limited controlled suckling, in order to evaluate at a long term the incidence on global productivity. At a commercial level the adoption of natural methods of reproductive control needs to achieve a compromise to compensate a potential decrease of growth, which could be solved through further research.

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