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PERRIER G., THEAU-CLÉMENT M. , JOUANNO M., DROUET J.P.

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REDUCTION OF THE GnRH DOSE AND INSEMINATED RABBIT DOE REPRODUCTIVE PERFORMANCE

PERRIER G.*, THEAU-CLÉMENT M. **, JOUANNO M.*, DROUET J.P.*

*Etablissement National d'Enseignement Supérieur Agronomique de Dijon
BP 87999, 21079 Dijon Cedex France. g.perrier@enesad.fr
**INRA, Station d'Amelioration Génétique des Animaux, BP 27, 31326 Castanet Tolosan Cedex, France

ABSTRACT

This experiment studied the effect of a reduced dose of a GnRH analogue (Receptal®) on reproductive performance of 120 rabbit does. One group received the usual dose of 0.2ml Receptal (0.8µg buserelin); the other group received 0.1 ml Receptal (0.4µg buserelin). A total of 768 inseminations were performed during the experiment. Overall, the fertility of the does receiving the lower dose was not significantly reduced (77.7 *vs.* 82.4 %). However, nulliparous does had fertility a decrease with the reduced dose (53.2 *vs.* 70.7%). The productivity at birth (11.0 *vs.* 10.9 kits born alive) and at weaning (9.2 *vs.* 9.3 kits weaned per litter) were not affected by the treatment. The importance of the physiological phase at insemination day (receptivity, parity, lactating or non- lactating does) is underlined here.

INTRODUCTION.

The use of artificial insemination (A.I.) combined with to batch management has become widely used in France. In 1997, TUDELA and MONTJOIE asserted that, in intensive rearing, half the population of does was bred through this method.

Ovulation must be induced for A.I. gonadoreline (Fertagyl, Laboratoire Intervet), a synthetic decapeptide with the same chemical structure as GnRH and buserelin (Receptal, Laboratoire Hoechst Roussel Vet), a nonapeptide with a structure similar to GnRH, are the most widely used products. The efficiency of these treatments is often deduced from fertility rates after insemination and rarely under real comparative conditions (MICHELMANN and PAUFLER, 1973; BATTAGLINI *et al.,*1982; RODRIGUEZ and UBILLA1988 ; ROCA and ALACE 1989). THEAU-CLEMENT *et al*. (1990) showed that 0.8 µg buserilin or 20 µg gonadoreline resulted in the same number of weaned kits per insemination. On rabbit farms, 0.2 ml Receptal (0.8 µg buserelin) is usually injected immediately after insemination.

Social pressure for limiting or eliminating regular use of synthetic products in rearing is increasing in Europe. This experiment fits into the prospect of limited hormone use in rabbit farms. The aim of this experiment was to study long term effects of the reduction of the Receptal injection dose (0.1 *vs*. 0.2 ml) on rabbit doe reproductive performance.

MATERIALS AND METHODS

One hundred twenty does sired by bucks of the A 2066 strain bred to does of the A 1077 strain were used. These were bred to 12 bucks from the Hyplus strain from March 1999 to January 2000.

The does were housed in individual flat-deck wire cages with plastic floors. They were fed *ad libitum* during the lactation period and given 150g commercial rabbit pellets a day the rest of the time. They were kept under artificial light 16 hours a day. The young does were first bred when they were 16 weeks old. Does were inseminated every 42 days (2 batches at 3 week intervals). Thirteen insemination series were made during the experiment.

Does found to be open 10 days after insemination were re-inseminated three weeks after the unsuccessful A.I. Does open twice in succession were culled. Primiparous and lactating does (except those submitted to a second A.I. because of a negative pregnancy diagnosis) were given 20 I.U. of PMSG 48h before insemination.

The herd was already productive when the experiment began. The does were homogeneously distributed in two groups according to their parity (nulliparous, primiparous and multiparous) and their physiological phase (lactating or not). Each doe remained in the same group throughout the experiment.

Semen was collected once a week. Ejaculates were selected only if free of urine, if the volume was equal to or greater than 0.5 ml, and overall motility equal to or greater than 7 (PETITJEAN, 1965). Semen was diluted 1:8 with Galap (IMV) at room temperature (18 to 20°C) within the five minutes following the sampling. After mixing and homogenising the ejaculates, semen was placed in 0.5 ml sheathed straws. These were kept horizontal at room temperature and used within two hours. On insemination day doe receptivity was tested by presentation to a buck. Does were considered receptive if they adopted the lordosis position. A.I. was performed by two operators who each inseminated the same number of does within each group and parity. The does were placed on their backs and inseminated with a curved pipette syringe introduced about 15 cm into the vagina. About sixty does were inseminated for each series, i.e. thirty in each group. Immediately after insemination, the does were given an intramuscular injection of 0.1 ml (group 1) or 0.2 ml (group 2) Receptal ®.

After each kindling series, litter sizes were homogenised after eliminating kits considered non-viable. Within each group, two homogenisation levels were applied - one for the primiparous does and one for rest of the does - based on the mean number of live kits within the subgroup. This was done 28 to 30 hours after the first kindling and after inducing the last kindling with 0.2 ml oxytocin. Weaning took place at 35 days.

Analysis of variance was carried out using the GLM procedure of SAS (SAS for Windows Version 6.12) with Receptal dose (0.1 or 0.2 ml), doe receptivity (R+ or R-), and physiological state at A.I. including lactation (L+ or L-) and parity (nulliparous L-, primiparous L+, primiparous L-, multiparous L+ and multiparous L-) as main effects. All two-way interactions were evaluated. Preliminary analyses indicated there was no effect of inseminator so this was eliminated from the final model. Dependent variables included fertility, litter size at birth (total kits born and kits born alive), litter size after adjustment and at weaning, and the number of kits weaned per A.I. Fertility was considered as a variable of Bernoulli (0-1).

RESULTS AND DISCUSSION

The analysis included 768 inseminations each with 27.5 to 82 x10^6 spermatozoa. The abortion rate did not differ between the groups with 4 in group 1 and 3 in group 2. The frequency of litters with no live kits was 1.5% for both groups. The use of ocytocin to induce kindling was the same in both groups (28 littering for group 1 and 30 for group 2).

Doe receptivity : There were 79.3% receptive does in group 1 and 81.9% in group 2. Primiparous does were less receptive than the multiparous (73.3 *vs.* 82.7%).

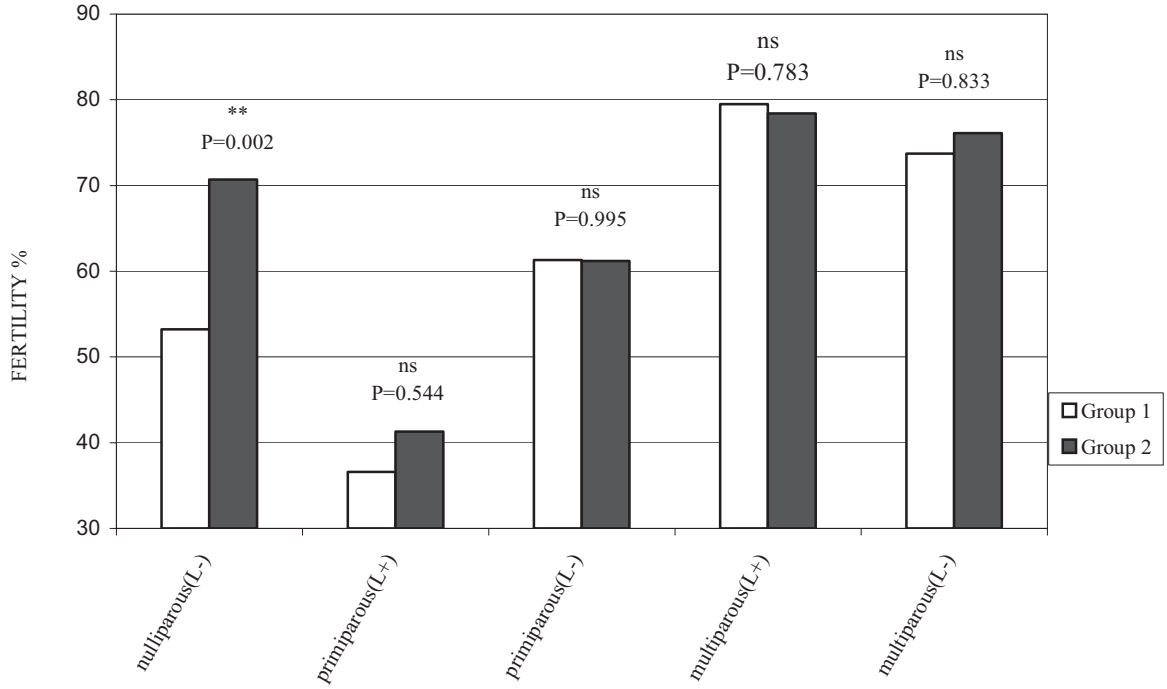

Figure 1 : Fertility in relationship to treatment group and physiological state

Fertility (% of does kindling) : Doe fertility was not significantly affected by reduction of the Receptal dose (Table 1). In fact, the nulliparous does in group 2 had higher fertility than their counterparts (70.7 *vs.* 53.2%, P<.01). For the other physiological states, doe performance was very similar in the two groups (Figure 1). The L+ primiparous does had the lowest fertility and the L- primiparous and nulliparous does had fertility less than or equal to than the multiparous does (Table 1).

Doe receptivity at insemination had an important (P<.001) effect on fertility with the R- does having a much lower fertility. This difference was seen regardless of parity. Nevertheless, deviations observed between R+ and R- varied greatly according to physiological state. These variations induced the only significant interaction observed. There was no difference in fertility between R+ and R- does of groups 1 and 2. This result is not in agreement with FOOTE and SIMKIN'S work (1993) that showed that, using Dutch rabbits, 0.4 µg buserelin was too small to elicit a good LH response and unlikely to provide a good ovulatory response. Nevertheless, these authors suggested that some genetic effects might be acting on buserelin sensitivity.

Prolificacy.: Receptal dose had no effect on total litter size, kits born live, or litter size at weaning. Nulliparous does had fewer kits born but primiparous (L+ or L-) does had litter sizes equal to multiparous does. Litter size born alive was lower for nulliparous does because of higher neonatal mortality. The R- does had fewer (P<.01) kits born alive than the R+ does. The

litter size adjustment at birth did not change the relationship between the two groups (10.9 *vs.* 11.0) although the difference was not significant.

Table 1 : Rabbit doe reproductive performance in relation to Receptal dose, physiological state, and receptivity at insemination day.

	N° A.I.	Fertility %	Total born	Born alive	Adjusted	Weaned / litter	Weaned / A.I.
			Arithmetic means				
- Group 1 (.1 ml)	*376*	77.7 *(41.7)*	11.6 *(2.8)*	11.0 *(3.1)*	10.9 *(1.3)*	9.2 *(2.9)*	6.7 *(4.8)*
- Group 2 (.2 ml)	*392*	82.4 *(38.1)*	11.7 *(2.6)*	10.9 *(3.3)*	10.8 *(1.6)*	9.3 *(2.7)*	7.2 *(4.6)*
			Sources of variation Least square means (s. e.)				
Receptal dose		ns	ns	ns	ns	ns	ns
- Group 1 (.1 ml)	*376*	60.9 *(2.8)*	11.4 *(0.3)*	10.3 *(0.4)*	10.9 *(0.2)*	9.6 *(0.3)*	5.5 *(0.3)*
- Group 2 (.2 ml)	*392*	65.5 *(2.9)*	11.3 *(0.3)*	10.3 *(0.4)*	11.0 *(0.2)*	9.5 *(0.3)*	5.8 *(0.4)*
Physiological state		***	***	***	***	***	***
- nulliparous(L-)	*168*	62.0^{b} *(3.4)*	9.5^{a} *(0.4)*	8.7^{a} *(0.5)*	9.1^{a} *(0.2)*	7.0^{a} *(0.4)*	4.2^{ab} *(0.4)*
- primiparous(L+)	*85*	39.0^{a} *(4.1)*	11.4^{b} *(0.7)*	9.8^{ab} *(0.8)*	11.2^{b} *(0.3)*	10.7^{b} *(0.7)*	3.8^{a} *(0.5)*
- primiparous(L-)	*46*	61.3^{b} *(6.6)*	11.6^{b} *(0.8)*	9.5^{ab} *(0.9)*	11.4^{b} *(0.4)*	10.1^{b} *(0.9)*	5.7^{bc} *(0.8)*
- multiparous(L+)	*427*	79.0^{c} *(2.3)*	11.9^{b} *(0.2)*	11.2^{bc} *(0.3)*	11.3^{b} *(0.1)*	9.6^{b} *(0.2)*	7.1^{c} *(0.3)*
- multiparous(L-)	*42*	74.9^{bc} *(5.7)*	12.5^{b} *(0.5)*	12.3^{c} *(0.6)*	11.8^{b} *(0.2)*	10.5^{b} (0.5)	7.6^{c} *(0.7)*
Receptivity		***	*	**	ns	ns	***
R -	*149*	41.8 *(3.6)*	10.9 *(0.5)*	9.3 *(0.6)*	11.0 *(0.2)*	9.6 *(0.5)*	3.7 *(0.4)*
R +	*619*	84.7 *(2.1)*	11.9 *(0.2)*	11.2 *(0.2)*	10.9 *(0.1)*	9.6 *(0.2)*	7.7 *(0.2)*
R^2		0.25	0.11	0.09	0.35	0.16	0.19

ns P>.05, * P<.05, ** P<.01, *** P<.001
Different letters in the same column indicate significant differences (P<.05)

The intra-parity standardisation for the does at first kindling caused a lower adjusted litter size than for the other parities. The adjustment for primiparous together with multiparous does led to an increase of adjusted litter size for the primiparous does. The number of kits weaned was the same in both groups. The number of kits weaned was less for the nulliparous does because they had smaller litters at kindling.

Global productivity (kits weaned/A.I.) : The number of kits weaned/A.I. not was not affected by the Receptal dose. The low performance of the nulliparous does (4.2 weaned kits per A.I.) is a result of the low fertility and litter size at birth. The number of kits weaned /A.I. was higher for the nulliparous does of group 2 than for those of group 1 (5.1 *vs.* 3.2 kits, P <.01) and their productivity was similar to that of the primiparous does. Primiparous does had a global productivity lower than the multiparous does because they were less fertile. In general, doe's receptivity greatly affects the weaned kits/A.I. (respectively 7.7 *vs.* 3.7 for R+ and R-) as it acts on fertility and prolificacy.

CONCLUSION

This experiment clearly reveals the factors necessary for a successful artificial insemination. Young nulliparous does are generally characterised as having low litter size (CASTELLINI, 1996). The primiparous does have some difficulties to simultaneously support their first lactation, the second gestation and their final growth (CHMITELIN et *al*.,1990; PARIGI-BINI and XICCATO, 1993; POUJARDIEU and THEAU-CLEMENT, 1995; FORTUN-LAMOTHE, 1998). These results confirm the importance of the doe's physiological state and receptivity at insemination

(THEAU-CLEMENT and ROUSTAN, 1992; FORTUN-LAMOTHE and BOLET, 1998; CASTELLINI, 1996; PERRIER *et al*., 1998).

Based on these results, we can suggest injecting nulliparous does with 0.2 ml Receptal and giving 0.1 ml Receptal to the other does. Injection of a small amount can present some risks if the material is not correctly injected into the muscle. These results should be confirmed in other rearing situations to confirm, in particular, the nulliparous response.

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