Limiting effects of uterine crowding on the number and weight of live pups at birth in hemiovariectomized and normal rabbit does

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Summary — Variation in the number, total weight, mean individual weight and minimum individual weight of live pups at first and second litter were studied in hemiovariectomized (H = 54 does) and normal (N = 55 does) rabbit does. The total number of implanted embryos in 25 unoperated (N) and 54 hemiovariectomized (H) does were recorded by laparoscopy on the 12th day of their second pregnancy.

The total number of implanted embryos was not statistically different between experimental groups (m ± SEM; H: 11.0 ± 0.4; N: 12.4 ± 0.5; P = 0.06). The number of total pups at birth (H: 7.7 ± 0.2 vs N: 9.8 ± 0.2; P < 0.01) was affected by hemiovariectomy even when analyzed as a constant total number of implanted embryos. The number of live pups (H: 7.0 ± 0.2 vs N: 9.0 ± 0.3; P < 0.01), the total weight of live pups (H: 395 ± 11 g vs N: 479 ± 12 g; P < 0.01), the mean litter weight (H: 58.9 ± 1.1 g vs N: 54.1 ± 0.7 g; P < 0.01) and the minimum individual weight of live pups at birth (H: 46.5 ± 1.4 g vs N: 42.1 ± 1.0 g; P < 0.05) were also affected by hemiovariectomy. The significance of hemiovariectomy effect disappeared when analyzed as a constant total number of pups at birth. A positive non-linear relationship between the total number of pups at birth and the total weight of live pups was detected. Negative non-linear relationships between the total number of pups at birth and both the mean litter weight and the minimum individual weight of live pups at birth were detected.

uterine crowding / hemiovariectomy / birth-weight / rabbit

Résumé — Effets limitants de la population utérine sur le nombre et le poids de lapereaux vivants à la naissance de lapines normales et hémiovariectomisées. La variation du nombre, du poids total, du poids moyen individuel et du poids minimal individuel de lapereaux vivants à la première et à la seconde portée a été étudiée sur des femelles hémiovariectomisées (H = 54 femelles) et normales (N = 55 femelles). Chez 25 femelles témoins non opérées (N) et 54 hémiovariectomisées (H) le nombre total d'embryons implantés est vérifié par coelioscopie le 12e j de leur second gestation. Le nombre total d'embryons implantés n'est pas différent entre les groupes expérimentaux m ± sem; H : 11.0 ± 0.4; N : 12.4 ± 0.5; P = 0.06). Le nombre de lapereaux totaux à la naissance (H : 7.7 ± 0.2 vs N : 9.8 ± 0.2; P < 0.01) est affecté par l'hémiovariectomie. Cet effet ne disparait pas lorsque les données sont rapportées à un nombre total d'embryons implantés constant. Le nombre de lapereaux vivants (H : 7.0 ± 0.2 vs N : 9.0 ± 0.3; P < 0.01), le poids total de lapereaux vivants (H : 395 ± 11 g vs N : 479 ± 12 g; P < 0.01), le poids moyen de la portée (H : 58.9 ± 1.1 g vs N : 54.1 ± 0.7 g;
P < 0.01) et le poids individuel minimal des lapereaux vivants à la naissance (H : 46,5 ± 1,4 g vs N : 42,1 ± 1,0 g; P < 0,05) ont été affectés par l'hémiovariectomie. La signification de ces effets disparaît lorsque les données sont rapportées à une taille de portée constante. Une relation positive non linéaire entre le nombre total de lapereaux à la naissance et le poids total de lapereaux vivants a été déteétée. On peut noter une relation négative non linéaire entre le nombre total de lapereaux à la naissance et le poids moyen de la portée ou le poids individuel minimal de lapereaux vivants à la naissance.

**population utérine / hémiovariectomie / poids à la naissance / lapin**

**INTRODUCTION**

In mammals, the major physiological components affecting prolificacy at birth are the number of oocytes ovulated and fertilized, the rate of embryonic survival, the physical and biochemical components of uterine capacity, the foeto-placental development and survival to term, as well as survival of the neonates as influenced by birthweight (Bazer et al., 1990).

In polytocous species it is possible to increase the number of implanted embryos by raising the ovulation rate and/or embryonic survival. However, the uterine capacity might become a limiting factor for the development and survival of an excessive number of conceptuses. In the rabbit, as the number of implantations rises, the distance between implantation sites and the size of placentae are correspondingly reduced (Adams, 1962). This provokes a lower foetal growth and/or a higher foetal mortality (Hafez, 1964). Uterine overcrowding can be induced experimentally in rabbits by hemiovariectomy since ovulation rate in the remaining ovary is similar to the total ovulation rate in normal does (Short et al., 1968) and transuterine migration of embryos does not occur in this species (Johnson, 1971). Accordingly, in these experimental animals, the number of implanted embryos can be expected to be slightly lower than the total number of implanted embryos in normal does.

The aim of this work was to study the effects of uterine crowding on the number and weight of live pups at birth in hemiovariectomized and normal rabbit does belonging to a strain selected for litter size.

**MATERIALS AND METHODS**

Rabbit does belonging to the SY strain (synthetic breed) were used. SY strain (Estany et al., 1988) has been selected for litter size at weaning. Fifty-four does were hemiovariectomized at 12-wk old (H) and 55 unoperated does were used as normal controls (N).

From 18 wk old onward, does were repeatedly presented to fertile bucks until they became pregnant. Nine of 10 d after their first farrowing, the does were presented to bucks until they achieved a second pregnancy.

During the 24 h following parturition, nests were inspected and the live pups were weighed. Stillborn pups were counted but not weighed because they had already experienced dehydration and possibly decomposition.

For each litter, the following data were recorded at birth: parity (first or second parturition); treatment (hemiovariectomized or normal); total number of pups (TN); number of live pups (LN); total weight of live pups (LW); mean weight of live pups (MW); minimum individual weight of live pups (OW).

Laparoscopy (Molina et al., 1987) was performed on 25 unoperated and 54 hemiovariectomized does on the twelfth day of their second pregnancy, noting the total number of embryos (TE) that were implanted in the uterus. Ovulation rates of these does were not recorded.
Statistical analysis

Data from laparoscopy

Effect of treatment (hemiovariectomy vs control) on the number of implanted embryos (TE): analysis of variance.

Effect of treatment on the total number of pups at birth (TN): analysis of variance and of variance-covariance, using the number of implanted embryos (TE) and TE^2 as covariates.

All parturitions

Effects of treatment, parity and their interactions on TN: analysis of variance.

Effects of treatment, parity and their interactions were studied on live pups at birth: number (LN), total weight (LW), mean weight (MW) and minimum individual weight (OW). These effects were analyzed by variance analysis and by variance-covariance analysis including as covariates the total number of pups at farrowing (TN) and TN^2.

Quadratic term (TE^2 or TN^2) was included in the analysis to test the non-linearity in the relationship between TE or TN and the dependent variables.

RESULTS

Pup number at implantation or at birth

The mean number of implanted embryos per female was not statistically different between hemiovariectomized does (H) and normal does (N) in their second pregnancy (m ± SEM; H: 11.0 ± 0.4; N: 12.4 ± 0.5; P = 0.06).

The total number of pups at birth was significantly different between hemiovariectomized rabbit does and normal does (H vs N: 7.7 ± 0.2 vs 9.8 ± 0.2; P < 0.01). Parity and interaction between treatment and parity were not significant. The significance of these differences remained when the analysis was restricted to the second litter of the does (after laparoscopy) for which the total number of implanted embryos (TE) and TE^2 were included as covariates. Only the covariate TE (coefficient: + 0.68; P < 0.05) was significant.

The lowest total litter size (TN) was from hemiovariectomized does, with 6 litters in which there were fewer than 4, the minimum value for TN observed in normal does. Conversely, the highest values for TN were from normal does, with 7 litters larger than 13, the maximum value observed for TN in hemiovariectomized does.

Live pups at birth: number and weight per litter

There was no significant interaction between treatment and parity in any of the following analysis.

Number of live pups per litter

This was not affected by parity. Differences between hemiovariectomized does and normal does (H: 7.0 ± 0.2 vs N: 9.0 ± 0.3; P < 0.01) disappeared when the covariates (TN, TN^2) were included in the model but only TN was significant (coefficient: + 1.2, P < 0.01).

Total weight of live pups per litter

No effect of parity was observed. The negative effect of hemiovariectomy (H: 395 ± 11 g vs N: 479 ± 12 g; P < 0.01) also disappeared when the covariates were included. Both covariates were significant (TN coefficient: + 65.4, P < 0.01; TN^2 coefficient: -2.1, P < 0.01).

Mean weight of live pups per litter

Differences between hemiovariectomized and normal does (H: 58.9 ± 1.1 g vs N:
54.1 ± 0.7 g; P < 0.01) disappeared when the covariates were included. On the contrary, the effect of parity (first: 55.3 ± 0.9 g vs second: 58.1 ± 1.1 g; P = 0.06) reached significance (P < 0.01) when the covariates were included. Both covariates were significant (TN coefficient: -6.1, P < 0.01; TN² coefficient: + 0.2, P < 0.01).

Minimum individual weight of live pups per litter

The differences between the H and N does groups (H: 46.5 ± 1.4 g vs N: 42.1 ± 1.0 g; P < 0.05) disappeared when the covariates were included. In this analysis both covariates were significant (TN coefficient: -8.6, P < 0.01; TN² coefficient: + 0.3, P < 0.01). Effect of parity was not detected.

Important differences were observed between the highest and the lowest minimum individual weights of live pups at birth (OWmax = 109 g vs OWmin = 22 g).

DISCUSSION

In hemiovariectomized does the level of crowding at initial stages of placentation (12 days pc) was high in the uterine horn ipsilateral compared with the remaining ovary. However, the experimentally-induced physical spacing restriction reduces the total number of foetuses that can be carried to term in this group. This effect was not totally explained by the initial number of implanted embryos. Thus, in agreement with Adams (1962), physical spacing in the uterus is a major limiting factor of foetal survival despite the remarkable distensibility and growth of the uterus during pregnancy. Moreover, when overcrowding is high (as in hemiovariectomized does) the survival to term could be affected for many conceptuses. Consequently, the litter size at birth could be reduced below the expected values (Hafez, 1964).

There are mechanisms in rabbit does which tend to minimize the frequency of litters with only 1 or 2 pups. They are related to the existence of a limit for both a lower value for ovulation rate (Garcia, 1982; Molina et al, 1987) and a minimum number of foetuses required for the maintenance of pregnancy (Hafez, 1968; Adams, 1970). Thus, the presence of only 1 or 2 placental foetuses does not prevent the activation of the luteolytic mechanism and consequently foetal loss occurs around d 21 postcoitum (Nowak et al, 1986).

The differences in the number of live pups at birth between treatments disappear when analyzed as constant number of total pups at birth. This suggests the absence of specific effect of treatment on perinatal losses.

When MW and OW were compared at constant number of total pups at birth, differences between hemiovariectomized does and normal does disappear. Thus, the increased foetal mortality determined by spacing restriction in hemiovariectomized does occurred mainly in the early placental stage. This would occur when overcrowding limits the growth of some placentae below a weight at which placenta and foetus cannot survive (Adams, 1962) but permits the remaining placentae and associated foetuses to grow normally until birth. The onset of the limiting effects of crowding on foetal growth is earlier in larger litters (Goss, 1978). In the rabbit, the effects of the number of surviving conceptuses is seen earlier in the placental weight (19 d) than in foetal weight (28 d) (García et al, 1983).

The difference observed between the lowest and the highest minimum individual weights of live pups at birth indicates the adaptability of foetal growth even in re-
stricted conditions during late pregnancy. In rabbit, the average number of runts at birth was 0.05 in litters of 6, 0.27 in litters of 10, and 0.64 in litters of 14 (Torres et al, 1986). McLaren and Michie (1960) working with mice showed that runts occurred at specific sites in the uterine horn. In these sites, the maternal blood supply to the placenta was poorer than elsewhere, but the resulting slow placental-foetal growth rate was still compatible with their survival to term. Thus, the weight of these runts expresses the minimum survival weight.

Non-linearity in the positive relationship between TN and LW and in the negative relationships between TN and both MW and OW was detected. This could be primary expressions of the limits proposed by Adams (1962) on raising the ponderal uterine capacity at full gestation and to the reduction in the foetal and placental development rates to a degree which should be compatible with their survival to term.

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