World Rabbit Science Association
Proceedings 10th World Rabbit Congress – September 3 - 6, 2012– Sharm El- Sheikh –Egypt, 367- 371

RELATIONSHIPS BETWEEN OVULATION RATE, LITTER SIZE AND PRENATAL SURVIVAL COMPONENTS IN RABBIT DOES

Ragab M.[1,2], Vicente J. S.[1], Lavara R.[1], Desantes, J.[1], Baselga, M.[1]

[1]Instituto de Ciencia y Tecnología Animal, Universidad Politécnica de Valencia,
Camino de Vera S/N, 46022, Valencia, Spain.
[2]Poultry Production Department, Fac. of Agriculture, Kafr El-Sheikh University, Egypt.
*Corresponding author: moramo@upvnet.upv.es

ABSTRACT

The aim of this study was to evaluate the relationships between ovulation rate (OR), implanted embryos (IE), total born (TB), number born alive (NBA) and embryo (ES), fetal (FS) and prenatal survival (PS) in females of high reproductive performance, resulting from a complete diallelic cross between four Spanish maternal lines of rabbits. Sixteen genetic types were studied (the four lines, and their twelve simple crosses). A total of 2025 does in their third, fourth or fifth gestations were subjected to a laparoscopy 11-12 days after mating. The previous traits were analysed with a model that included for all the traits, the genetic group-farm-year-season combination (GFYS), the parity order and the lactation state. For all the traits, excepted OR, the linear and quadratic effects of a covariate were considered. The covariate OR was included for the analysis of the other traits; IE for TB, NBA and FS; and NBA for TB for NBA. The general means obtained for the traits were: 15.2 ova for OR, 13.4 embryos for IE, 11.1 rabbits for TB, 10.2 rabbits for NBA, 87.9% for ES, 84.9% for FS and 74.0% for PS. None effects of parity order and lactation stages were observed, but the GFYS effect was significant. It is remarkable that survival rates obtained were high, with prenatal survival higher than 70% and proportional distribution of losses before and after 12th day of gestation. A negative curvilinear and significant relationship was found between ovulation rate and litter size components, affecting in the same sense the survival rates, but only the coefficients of regression of the quadratic term were significant for ES and PS. Moreover, the relationship of the covariate implanted embryos with TB, NBA and FS, and also the covariate TB with NBA were also significant and negative. This could be considered as a primary expression of the uterine capacity limits. The significant regression coefficients of the linear term ranged between 1.20 and 1.36, and the corresponding coefficients of the quadratic term, between -0.03 and -0.02 for IE, TB and TBA. In spite of significance of the quadratic coefficients, they might be not relevant because it closed to zero. No effects of parity order and lactation status were observed. A negative curvilinear and significant relationship was found between ovulation rate and the rest of litter size components, affecting in the same sense the survival rates.

Key words: Rabbits, laparoscopy, litter size components, prenatal survival, quadratic regression.

INTRODUCTION

In multiparous species, the major limiting factor in prenatal growth could be the uterus, despite its remarkable extensibility and growth during pregnancy. Then a physical spacing restriction in the uterine horns could reduce the weight of foetuses carried to term and increase the mortality at the end of gestation, at birth and in the first days of postnatal life. The higher the number of foetuses at birth, the greater the total weight of litter. However, the relationship between these traits does not appear to be linear (Salmon-Legagneur, 1968; Wilmut *et al*, 1990, García-Ximénez and Vicente, 1993 and Vicente *et al.,* 1995), which implies a progressive reduction of individual weight of young at birth when the number of foetuses increases.

In rabbits, there are three critical moments for foetal survival, the first between days 8 and 17 of gestation, when the hemochorial placenta has finished its development and the nutrition of the foetus begins to be controlled by the placenta (Adams, 1960) and it does not seem to be affected by uterine capacity. A second peak of mortality occurs between days 17 and 24 *post coitus*, corresponding with the period of uterine enlargement, when the tension on the spherical *conceptus* is at a maximum and blood flow through the maternal vessels of the uterus decreases (Hafez and Tsutsumi, 1966; Argente *et al*., 2003 and 2008). And the third can take place in the last week of gestation, when the energy requirements for the foetal growth increase rapidly whereas the feed ingestion declines in the days preceding the birth (Fortun-Lamothe, 2006).

Adams (1962) proposed the existence of limits for the reduction in foetal and placental development rates of live young rabbits at birth. Duncan (1969) observed that the more foetuses in the uterine horn, the lower the maternal blood flow to the placenta, which was more likely to result in the presence of runts. Breuer and Claussen (1977), García-Ximénez and Vicente (1993) and Vicente *et al*. (1995) observed a negative correlation between the foetal and born alive weight and the litter size, which further limits the birth weight of each member of a multiple birth (Widdowson, 1976, Vicente *et al*., 1995). So, if birth weight is lower than the optimum weight, energy reserves and thermoregulatory capacity are reduced and perinatal mortality is increased.

The aim of this study was evaluate the relationships between ovulation rate, implanted embryos, total born, number of born alive rabbits and embryo, fetal and prenatal survival in females of high reproductive performance.

MATERIALS AND METHODS

Animals and management

A complete diallelic cross involving four maternal lines of rabbits (A, V, H, and LP, selected for litter size at weaning; described previously by Ragab and Baselga, 2011) was carried out. Their current generation of selection was 41st, 37th, 20th and 7th, respectively. Data were collected from January 2009 to April 2011. The experimental work was carried out in four Spanish farms with a total of 2260 cages available for breeding animals. The farms were located in León (farm 1: 800 does), Castellón (farm 2: 800 does), Tarragona (farm 3: 300 does) and the farm of Universidad Politécnica de Valencia (UPV, farm 4: 360 does). The genetic groups involved in the experiment were the four lines (AA, VV, HH and LL) and the twelve single crosses (AV, VA. AH, HA, AL, LA, VH, HV, VL, LV, HL and LH, the first letter refers to the sire line, and the second to the dam line; L is used to identify the LP line). Farms 1 and 2 raised all the crossbreds and purebred VV line animals; the farm 3 raised females of the groups VV and HH and the farm 4 housed VV, AA and LL animals. The group VV was present in all the farms to connect data between farms.
Management was slightly different between farms. In farm 1 and 2 a single batch every 42 days was applied using artificial insemination, while in farms 3 and 4, natural mating was practiced each week (6 batches). In all farms, the first mating of does and bucks was around 18 weeks of age. Rabbits were fed *ad libitum* with a standard commercial pelleted diet and keep under a constant photoperiod of 16L:8D. In farms 1 and 2, 60-62 hours before the insemination all does were injected with eCG (20 eCG I.U. by doe). The does were inseminated with heterospermic pools of a paternal line (line R, Estany *et al*., 1992). Females were inseminated with 0.5 ml diluted semen and ovulation was induced by an intramuscular injection of 1µg of busereline acetate. Semen doses were elaborated 16 hours before according to following quality criteria: motility rate higher than 70%, abnormal sperm lower than 15% and a concentration around 20-40 million spermatozoa per ml. The males in farms 3 and 4 were from the same lines as the females.

Traits and statistical analyses

The studied traits were ovulation rate (OR; estimated as the number of corpora lutea in both ovaries), number of implanted embryos (IE; measured as the total number of implantation sites), total born (TB; measured as the total number of kits born), number born alive (NBA; measured as the number of kits born alive), embryo survival (ES; estimated as IE / OR), fetal survival (FS; estimated as TB / IE), and prenatal survival (PS; estimated as TB / OR). These records were obtained from does in their third, fourth or fifth gestations that were subjected to a laparoscopy 11-12 days after mating according to the technique described by Santacreu *et al*. (1990). A total of 2025 does, from the sixteen genetic types were subjected to laparoscopy.

Different models were fitted depending on the trait and the trait that was used as a covariate. For OR the following model was used:

$$Y_{ijk} = GFYS_i + OP_j + L_k + e_{ijk} \qquad \text{(Model 1)}$$

where, Y_{ijkl} is a record of the trait to be analysed,
$GFYS_i$ is the fixed effect of the genetic group-farm-year-season combination (129 levels); OP_j is the parity order (3 levels: 3, 4 and 5); L_k is the effect of lactation state of the doe (2 levels: lactating does and not lactating does at mating time), and e_{ijk} is the residual of the model.

To analyse IE, TB, NBA, ES, FS and PS a model with the same fixed effects that model 1, adding a linear and quadratic terms of a covariate X. The model was,

$$Y_{ijkl} = GFYS_i + OP_j + L_k + \beta (X_{ijkl}) + \gamma (X_{ijkl})^2 + e_{ijkl} \qquad \text{(Model 2)}$$

where, β and γ are regression coefficients on X and X^2. The covariate X was, OR for all afore mentioned traits; IE for TB, NBA and FS; and TB for NBA. The effect of natural and artificial insemination were included in genetic group-farm-year-season.
The models were solved and least squares means for fixed effects was determined using option of the GLM procedure of SAS (version 9.1.3; SAS Institute, 2002). Significance level $\alpha = 0.05$

RESULTS AND DISCUSSION

Table 1: Litter size components per parity order and lactation stage (lsmeans (standard errors).

Parity order	N	OR	IE	TB	NBA	ES	FS	PS
3	991	15.1(0.12)	13.5(0.10)	11.2(0.14)	10.1(0.09)	88.5(0.69)	84.9(0.84)	74.5(0.92)
4	745	15.3(0.12)	13.4(0.11)	11.1(0.15)	10.2(0.09)	87.8(0.73)	84.7(0.90)	73.7(0.99)
5	289	15.2(0.18)	13.3(0.16)	11.1(0.22)	10.1(0.14)	87.7(1.07)	85.0(0.32)	73.7(1.46)
Lactation status								
Lactating	1591	15.3(0.09)	13.2(0.08)	10.9(0.11)	10.2(0.07)	87.1(0.56)	84.2(0.68)	72.5(0.75)
Non lactating	434	15.1(0.15)	13.5(0.13)	11.3(0.18)*	10.1(0.11)	88.8(0.89)	85.5(1.10)	75.5(1.21)

N: the records number; OR: ovulation rate, IE: number of implanted embryos, TB: total born, NBA: number born alive, ES: embryo survival rate, FS: foetal survival rate and PS: prenatal survival.

No effects of parity order and lactation status were observed (Table 1), while GFYS effect (genetic group-farm-year-season) was significant. It is remarkable that survival rates were higher than those obtained by other authors (Santacreu *et al*., 2005; Ibáñez-Escriche *et al*., 2006; Laborda *et al*., 2011, 2012), with prenatal survival higher than 70% with a similar proportional distribution of losses before and after 12th day of gestation. This is good indicator of the high level of reproductive performances of maternal lines and their dialell crossbreds.

A negative curvilinear and significant relationship was found between ovulation rate and the rest of litter size components, affecting in the same sense the survival rates (Table 2), but only the coefficients of regression of the quadratic term were significant for ES and PS. Moreover, the relationship of the covariate implanted embryos with TB, NBA and FS and PS and the covariate TB and NBA were also significant and negative. This could be considered as a primary expression of uterine capacity to maintain viable foetuses. García-Ximénez and Vicente (1993) showed only a negative curvilinear relationship between the total born and both the mean litter weight and the minimum individual weight of born alive at birth. Vicente *et al.* (1995) observed a negative relationship between born alive and total born in three lines selected by different criteria, being the regression coefficients of curvilinear term different from paternal and maternal lines. They suggested further limits to the weight of each member of a multiple birth raising prenatal and perinatal losses.

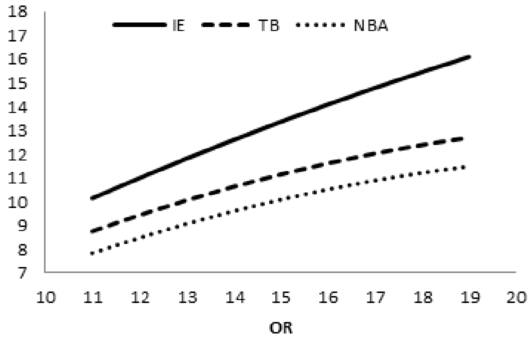

Fig 1. Relationship between the OR and IE, TB and NBA

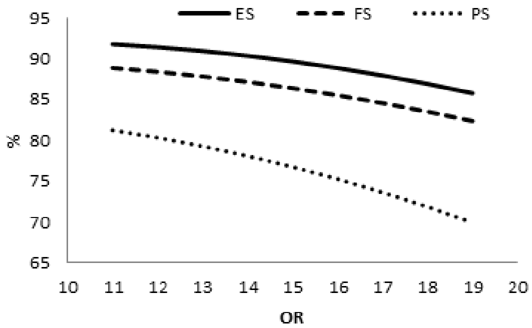

Fig 2. Relationship between OR and ES, FS and PS

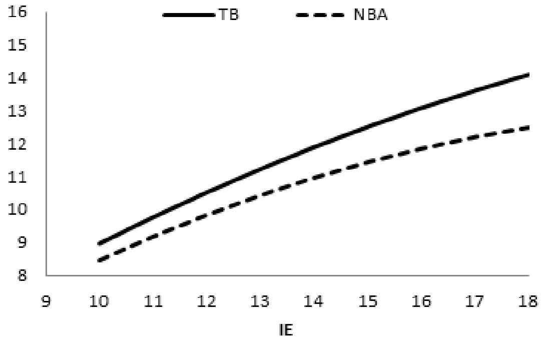

Fig3. Relationship between IE and TB and NBA

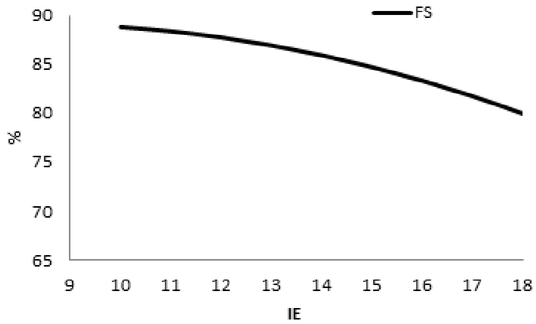

Fig 4. Relationship between IE and FS

Table 2: Linear and quadratic regression coefficients (standard error) of ovulation rate (OR), number of implanted embryos (IE) and total born (TB) for litter size components and survival rates.

	OR	OR^2	IE	IE^2	TB	TB^2
IE	1.20(0.13)*	-0.015(0.003)*				
TB	1.29(0.17)*	-0.026(0.005)*	1.27(0.08)*	-0.023(0.003)*		
NBA	1.20(0.20)*	-0.027(0.006)*	1.36(0.10)*	-0.030(0.004)*	1.3(0.06)*	-0.019(0.002)*
ES	0.92(0.84)	-0.056(0.025)*				
FS	0.62(1.03)	-0.048(0.031)	1.57(0.62)*	-0.096(0.024)*		
PS	0.79(1.13)	-0.073(0.034)*				

* Significant, α=0.05. NBA: number born alive, ES: embryo survival rate, FS: foetal survival rate, PS: prenatal survival.

Argente *et al.* (2008) observed that a reduction of the uterine space available was linearly associated with a reduction of the fetal survival by means of a limitation in the development of the maternal placenta and a reduction in the number of blood vessels arriving at each implantation site. So, quadratic regression coefficients of maternal-placenta length per foetus were higher in unilaterally ovariectomized than in intact does. Figures 1 to 4 represent the significant curvilinear relationships among each trait and one of the previous components of litter size. These figures show that in spite of significance of the quadratic coefficients, they might be not relevant to determine curvilinearity because it was closed to zero. It found to be not different from zero in intact does (Blasco *et al.,* 1996). Retrospective and further studies of the relationship between foetal survival and litter size at birth could be interesting in the maternal lines used for the diallel cross.

CONCLUSION

No effects of parity order and lactation status were observed. A negative curvilinear and significant relationship was found between ovulation rate and the rest of litter size components, affecting in the same sense the survival rates.

ACKNOWLEDEGEMENTS

The authors are thankful to the projects CICYT AGL2008-03274 and Prometeo 2009/125.

REFERENCES

Adams C. E. 1960. Prenatal mortality in the rabbit *Oryctolagus cuniculus*. *J. Reprod. Fertil*. I:36-44.

Adams C. E. 1962. Studies on prenatal mortality in the rabbit, oryctolagus cuniculus: the effect of transferring varying numbers of eggs. *J. Endocrinol*, 24, 471-490.

Argente M. J., Santacreu M. A., Climent A., Blasco A. 2003. Relationships between uterine and fetal traits in rabbits selected on uterine capacity. *J. Anim. Sci*., 81: 1265-1273.

Argente M. J., Santacreu M. A., Climent A., Blasco A. 2008. Effect of intrauterine crowding on available uterine space per fetus in rabbits. *Livest. Sci*., 114:211-219.

Breuer, H.W., Claussen V. 1977. Correlation of birth weight and crown-rump to the number of implantations and litter size in rabbits. *Anat. Embryol*., 151: 91-95

Duncan, S. L. B. 1969. The partition of uterine blood flow in the pregnant rabbit. *J. Physiol., Lond.* 204,. 421-433.

Estany J., Camacho J., Baselga M., Blasco A. 1992. Selection response of growth rate in rabbits for meat production. *Genet. Sel. Evol*., 24, 527–537.

Fortun-Lamothe L. 2006. Energy balance and reproductive performance in rabbit does. *Anim. Reprod. Sci.*, 93:1-15.

Garcia-Ximénez, F., Vicente J. S. 1993. Limiting effects of uterine crowding on the number and weight of live pups at birth in hemiovariectomized and normal rabbit does. *Reprod. Nutr. Dev*., 33: 69-73.

Hafez, E. S. E., Tsutsumi Y. 1966. Changes in endometrial vascularity during implantation and pregnancy in the rabbit. *Am. J. Anat*., 118: 249-282.

Ibañez-Escriche, N., M. A. Santacreu, M. Martinez, A. Climent, A. Blasco. 2006. Selection for ovulation rate in rabbits. *Livest. Sci., 101:126-133.*

Laborda, P., M. L. Mocé, M. A. Santacreu, A. Blasco. 2011. Selection for ovulation rate in rabbits: I. genetic parameters, direct response and correlated response on litter size. *J. Anim. Sci.,*10:2981-2987*.*

Laborda, P., M. L. Mocé, A. Blasco, M. A.Santacreu. 2012. Selection for ovulation rate in rabbits: genetic parameters and correlated responses on survival rates. *J. Anim. Sci., 2:439-446*.

Ragab, M., Baselga M. 2011. A comparison of reproductive traits of four maternal lines of rabbits selected for litter size at weaning and founded on different criteria. *Livest. Sci. 136, 201-206.*

Salmon-Legagneur, E. 1968. Prenatal development in the pig and some other muhiparous animals. P. 158-91. In G. A. Lodge and G. E. Lamming. (Ed.) *Growth and Development of Mammals.* Plenum Press, New York.

Santacreu, M. A., Viudes M. P., Blasco A. 1990. Evaluation par coelioscopie des corps jaunes et des embryons. Influence sur la taille de portée chez la lapine. *Reprod. Nutr. Dev.* 30:583–588.

Santacreu, M. A., M. L. Mocé, A. Climent, A. Blasco. 2005. Divergent selection for uterine capacity in rabbits. II. Correlated response on litter size and its components estimated with a cryopreserved control population*. J. Anim. Sci., 83:2303-2307.*

SAS. 2002. SAS/STAT User's Guide (Release 9.1.3). *SAS Inst. Inc., Cary NC, USA*.

Vicente J.S., Garcia-Ximénez F., Viudes-de-Castro M.P. 1995. Neonatal performances in 3 lines of rabbit (litter sizes, litter and individual weights). *Ann. Zootech.* 44: 255-261.

Widdowson E.M. 1986. Environmental control of growth: the maternal environment, *In: Lister D., Rhodes DN., Fowler VR., Fuller MS. (Eds). Meat animals, growth and productivity, Plenum Press, New York,* 273-284.

Wilmut I., Sales D.I., Ashworth C.J. 1986. Maternal and embryonic factors associated with prenatal loss in mammals. *J. Reprod. Fert*., 76: 851-864.