

Proceedings of the



4-7 July **2000** – Valencia Spain

These proceedings were printed as a special issue of *WORLD RABBIT SCIENCE*, the journal of the World Rabbit Science Association, Volume 8, supplement 1

**ISSN reference of this on line version is 2308-1910**

*(ISSN for all the on-line versions of the proceedings of the successive World Rabbit Congresses)*

**LAVARA R., MOCE E., ANDREU E., PASCUAL J.J., CERVERA C.,  
VIUDES-DE-CASTRO M.P., VICENTE J.S.**

**EFFECTS OF ENVIRONMENTAL TEMPERATURE  
AND VITAMIN SUPPLEMENTS  
ON SEMINAL PARAMETERS FROM A RABBIT LINE  
SELECTED BY HIGH GROWTH RATE**

Volume A, pages 167-171

# EFFECTS OF ENVIRONMENTAL TEMPERATURE AND VITAMIN SUPPLEMENTS ON SEMINAL PARAMETERS FROM A RABBIT LINE SELECTED BY HIGH GROWTH RATE

LAVARA R.\*, MOCE E.\*; ANDREU E., PASCUAL J.J.\*, CERVERA C.\*, VIUDES-DE-CASTRO M.P.\*\*; VICENTE J.S.\*

\*Departamento de Ciencia Animal, Universidad Politécnica de Valencia, 46071-Valencia, Spain.

\*\*División de Producción Animal, Universidad Miguel Hernández, 03312-Orihuela, Spain.  
rlavara@dca.upv.es

## ABSTRACT

Thirty four males born in spring were used to study the effect of vitamin supplements and the environmental temperature on seminal parameters from a rabbit line selected by high growth. Two pelleted diets were evaluated: a typical commercial diet (diet C) containing a vitamin/mineral mixture with 8400, 20 and 670 UI of vitamins A, E and D3 respectively; and a vitamin supplemented diet (diet V) from diet C, containing 11000, 50 and 800 UI of vitamins A, E and D3 in its mixture respectively. Two ejaculates per male were collected each week using an artificial vagina. Non significant differences were observed in collection rate (89% and 87%, V diet and C diet, respectively) and age which developed the mate behaviour (160±2 and 156±2 days of age, V diet and C diet respectively). The slight increase of the temperature during summer ( $T_{\max}^a$ : 26°C) affected negatively the feed intake of rabbits (-94 g/day) and the sperm production ( $-100 \cdot 10^6$  spermatozoa) of semen collected two months after. There were not significant differences in all the semen traits evaluated, showing similar semen volume and total sperm production with both diets. In conclusion, the supplementation of commercial diets with a higher content on vitamins A, E and D3 did not seem to improve collection rate, age of initial mate behaviour or semen quality and production.

## INTRODUCTION

Artificial insemination is valuable for rabbit producers because males selected on the basis of desired traits of rapid and efficient growth could be used to inseminate large population of females. In addition, the population of males as well as reproductive management time can be reduced. However, because of the relatively low sperm count of this species (200-600 millions) and the ordinary number of spermatozoa used per insemination (16-30 millions) the number of insemination per week and male is reduced to 10 or 20.

Several studies have been realised to increase the number of doses and quality of sperm produced by male, reducing the number of sperm in insemination doses (Viudes *et al.*, 1997) or improving male management (Theau-Clément *et al.*, 1995; Rebollar y Alvarino, 1997; Mocé *et al.*, 1999). However, only a few studies have been done on nutritional requirements of male and their relation with sperm quality and production (Castellini *et al.*, 1999).

As it is well known in other species, the use of some natural antioxidants could improve the quality of semen, mainly due to their role in the protection of the sperm membrane integrity, in view of the high susceptibility to peroxidation of their unsaturated fatty acids.

The aim of this study was to evaluate the effect of vitamin supplementation on some semen traits from rabbit males belonging to a line selected by growth rate housed under farm conditions.

## MATERIAL AND METHODS

### Animals.

Thirty four males from line R born in spring were used in this experiment carried out from June to October. Line R was selected on the basis of growth rate from weaning to slaughter (28-63 days of age, Estany *et al.*, 1992). Males with 9-10 weeks of age were assigned randomly to diet groups and were housed in individual cages with light alternating on a cycle of 16 light hours and 8 dark hours. Environmental temperature of experimental period were noted.

### Diets

Two pelleted diets were formulated: a typical commercial diet (diet C) with 179 g CP kg<sup>-1</sup> DM and 12.0 MJ DE kg<sup>-1</sup> DM containing a vitamin/mineral mixture, respectively, 8400, 20 and 670 UI of vitamins A, E and D<sub>3</sub>; and a vitamin supplemented diet (diet V) from diet C, containing 11000, 50 and 800 UI of vitamins A, E and D<sub>3</sub> in its mixture, respectively. They were offered *ad libitum* during all the experimental period (9-10 weeks age to 28-32 weeks age).

### Semen collection and evaluation

Males were stimulated to mate at 18 weeks of age with receptive does. Two ejaculates per male were collected each week using an artificial vagina. Data from the 10 first weeks of sexual activity was recorded. The following measurements were taken from semen:

- a) Volume of semen was measured in a graduated conical tube.
- b) Concentration of sperm per mL was calculated with a hemocytometer.
- c) Acrosomal integrity and abnormal sperm: spermatozoa were fixed with glutaraldehyde 2% in Dulbecco's phosphate buffered saline (Pursel and Johnson, 1974) and the proportion of either sperm with normal intact acrosome and abnormal sperm morphologically was estimated using interference contrast optics at a magnification of x750.

### Statistical analysis

A chi-squared with correction Yate's was used to analyse the collection rate (number of ejaculates/number of collections –ejaculates, ejaculates with urine, urine) between diets.

Age of male at the first mate and semen parameters were analysed by variance analysis, using a mixed procedure (PROC MIXED) of SAS (Statistical Analysis System Institute, 1996) and according to a repeated measures design that take into account the variation between animals and covariation within them. The model included like fixed effects the diet (2 levels) and the week (10 levels), using the present temperature and the temperature of two months previous to the collection as covariate. Covariance structures of mixed procedure were objectively compared using the most severe criteria (Schwarz Bayesian criterion), as suggested by Littell *et al.* (1998).

## RESULTS AND DISCUSSION

Five-hundred and seventy-seven ejaculates out of 644 semen collection were obtained, only a 12% (77) of collection were urine o semen with urine. Non significant differences were observed in collection rate (89% and 87%, V diet and C diet, respectively) and age which developed the mate behaviour (160±2 and 156±2 days of age, V diet and C diet respectively).

Data in Table 1 show that there were not significant differences in all the semen traits evaluated, showing similar semen volume (mean 1.19 ml) and total sperm production (mean 208·10<sup>6</sup>) with both diets. The vitamin supplementation of diet did not affect either acrosoma integrity (mean 81.9%) or total sperm abnormalities (12.8 and 9.0 for C and V diets, respectively).

**Table 1.** Effect of dietary vitamin supplementation on the semen characteristics.

	Diets		SE	P
	C	V		
Semen volume (ml)	1.248	1.127	0.0863	0.2779
Sperm production (10 <sup>6</sup> )	210.1	206.6	20.62	0.8965
Sperm concentration (10 <sup>6</sup> /ml)	193.1	192.4	23.00	0.9801
Acrosoma integrity (%)	83.62	80.11	2.296	0.2340
Sperm abnormalities (%)	12.82	8.99	2.257	0.2164

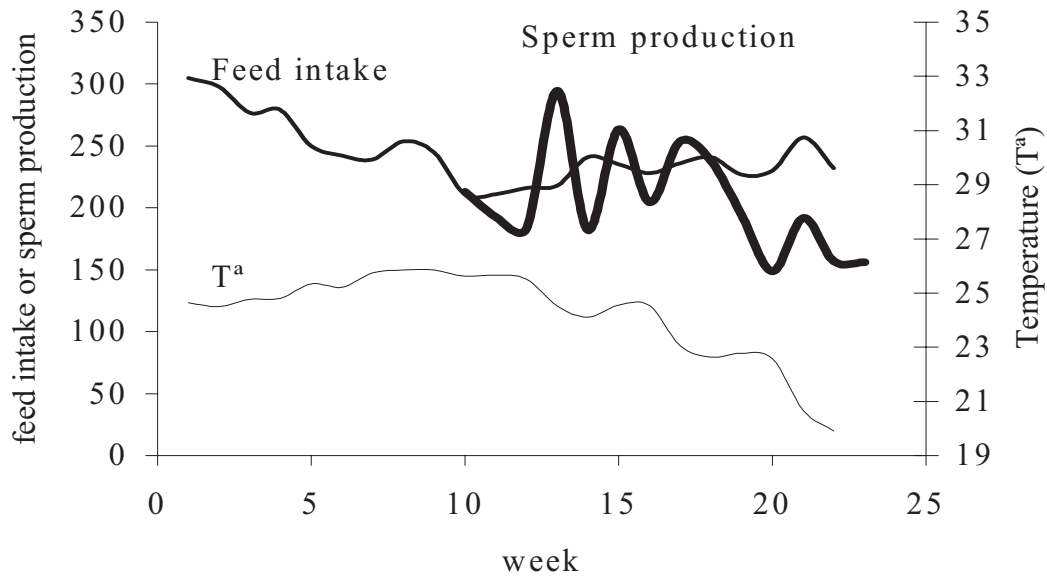
SE: standard error

P: statistical significance

Although the environment was controlled, the slight increase of the temperature (T<sup>a</sup><sub>max</sub>: 26°C) affected negatively the feed intake of rabbits during summer (-94 g/day), and consequently also affected the sperm production (-100 x10<sup>6</sup> spermatozoa) of semen collected two months after (figure 1). The negative effect of temperature on reproductive performance is well known. The duration of spermatogenesis in rabbit is about 42 days so, the high temperatures of August (26-27°C) affected semen production of male in October.

The results of the present work indicated that the supplementation of commercial diets with a higher content on vitamins A, E and D3 do not seem to improve collection rate, age of initial mate behaviour or semen quality and production. Similar results have been also found by other authors (El-Masry *et al.*, 1994; Castellini *et al.*, 1999), that did not show any effect of vitamin supplementation on the semen production (volume or concentration) and morphological parameters of quality (abnormalities). Castellini *et al.* (1999) showed that the addition of vitamin E alone reduced some kinetic parameters (progressive speed and linearity), but these were increased by the combination of vitamins E and C. However, these improvement of kinetic parameters were not related with fertility rate of fresh semen. If vitamin supplement affects other seminal characteristics as time of conservation or freeze ability, it should be studied in order to management of semen production.

**Figure 1.** Effect of maximum temperature (°C) on the feed intake (g/day) and sperm production ( $10^6$  spermatozoa) of males throughout the experimental period.



#### ACKNOWLEDGEMENTS

This study was supported by CICYT project n°AGF98-470-C02-01 and Nanta S.A. We thank CUNIZAR S.A. for helpful collaboration.

#### REFERENCES

- CASTELLINI, C., LATTAIOLI, P. AND BERNARDINI, M., 1999. Effect of dietary supplementation with  $\alpha$ -tocopheryl acetate and ascorbic acid on qualitative characteristics and fertilizing ability of rabbit semen. *World Rabbit Science*, 7: 217-220.
- EL-MASRY, K.A., NASR, A.S. AND KAMAL, T.H., 1994. Influences of season and dietary supplementation with selenium and vitamin E or zinc on some blood constituents and semen quality of New Zealand white rabbit males. *World Rabbit Science*, 2: 79-86.
- 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.
- LITTELL, R.C., HENRY, P.R. AND AMMERMAN, C.B., 1998. Statistical Analysis of Repeated Measures Data Using SAS procedures. *Journal of Animal Science*, 76: 1216-1231.
- MOCE, E., LAVARA, R., VICENTE, J.S., 1999. Efecto del manejo reproductivo de los machos de una línea de elevada velocidad de crecimiento sobre los parámetros seminales. II Congreso Ibérico de Reproducción Animal, Lugo- España. 480-482.
- PURSEL, V.G., JOHNSON, L.A., 1974. Glutaraldehyde fixation of boar spermatozoa for acrosome evaluation. *Theriogenology*, 2: 63-68.

- REBOLLAR, P.G., ALVARIÑO, J.M.R., 1997. Efecto de un tratamiento continuado con gonadorelina y hCG sobre parámetros seminales de conejos jóvenes. ITEA, 18 (II): 460-462.
- STATISTICAL ANALYSIS SYSTEMS INSTITUTE. 1996. User's guide statistics. Statistical Analysis System Institute Inc., Cary.
- THEAU-CLÉMENT, M., MICHEL, N., ESPARBIÉ, J., BOLET, G., 1995. Effects of artificial photoperiods on sexual behaviour and sperm output in the rabbit. *Animal Science*, 60: 143-149.
- VIUDES DE CASTRO, M.P., VICENTE, J.S., 1997. Effect of sperm count on the fertility and prolificity rates of meat rabbits *Anim. Reprod. Sci.*, 46. 313-319.