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**GUTIÉRREZ I., GARCIA REBOLLAR P.,
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EFFECT OF SUPPLEMENTATION WITH ANIMAL PLASMA AND ANTIBIOTICS ON JEJUNAL MORPHOLOGY OF EARLY-WEANED RABBITS

GUTIÉRREZ I., GARCIA REBOLLAR P., CARABAÑO R., DE BLAS J.C.

Departamento de Producción Animal, Universidad Politécnica de Madrid
E.T.S.I. Agrónomos, 28040, Madrid, Spain
cdeblas@pan.etsia.upm.es

ABSTRACT

Forty-two early-weaned (25 days) New Zealand x Californian rabbits were used to evaluate inclusion of a 4% of animal plasma (Appetein[®], APC Europe (SPAIN)) in substitution of soybean meal, and antibiotic supplementation (a mixture of 100 ppm of Bacitracine and 60 ppm of Apramicine) in four iso-nutritive starter diets. Type of diet did not affect feed intake or growth rate in the period from 25 to 35 days of age. Both animal plasma inclusion and antibiotic supplementation were effective in reducing mortality caused by enteritis in this period. Inclusion of animal plasma had only a significant effect on intestinal morphology when no antibiotics were added to the diet. In this case, a 4% addition of animal plasma improved ($P = 0.08$) villus height of jejunal (by 31.2%) and depth of crypts (by 14.4%) in 35-days old animals.

INTRODUCTION

Early-weaning at 25 days of age might have an interest to enhance reproductive performance of intensively-bred rabbit does. However, early weaning also implies an increase of nutritional and immunological problems (De Blas et al., 1999). Proper design of starter diets should take into account not only its effects on young rabbit performance, but also on the intestinal health.

The gastrointestinal tract undergoes many changes after weaning. In piglets, the villi are much shorter and the crypts are generally deeper with respect to the suckling period (Alle and Touchette, 1999). Inclusion of animal plasma in starter diets for piglets has led to an increase of weight gain and of villus height (Touchette et al., 1997).

Previous work (Gutiérrez et al., 2000) showed that animal plasma is a palatable and highly digestible source of protein for early weaned rabbits. The objective of this study was to determine the effect of inclusion of animal plasma (Appetein[®], APC Europe, SPAIN) and antibiotics on the villus height and the depth of crypts of jejunal epithelial cells in 35 day old rabbits weaned at 25 days of age.

MATERIALS AND METHODS

Diets

There were four experimental treatments. Treatments T1 and T2 had 0 and 4% of animal plasma added. Treatments T3 and T4 were based on treatments T1 and T2, respectively, adding a mixture of 100 ppm of bacitracine and 60 ppm of apramicine.

The different diets were formulated to meet or exceed all the essential nutrient requirements of growing rabbits (De Blas and Mateos, 1998). Their ingredient, chemical composition and calculated nutritive value are shown in Tables 1, 2 and 3, respectively. In order to formulate isonutritive diets, soybean meal and soybean hulls were partially replaced by animal plasma, barley, beet pulp and sepiolite.

Table 1. Ingredient composition of experimental diets, (%)

<i>Ingredient (%)</i>	Diets	
	1 & 3	2 & 4
Animal plasma	0.0	4.0
Wheat	16.4	16.4
Lard	2.5	2.5
Gluten 20	10.0	10.0
Wheat bran	20.0	20.0
Sunflower hulls	5.0	5.0
Lucerne hay	23.9	23.9
Soybean meal	8.9	0.0
Soybean hulls	10.0	7.7
Barley	0.0	0.47
Sugar-beet pulp	0.0	5.5
Sepiolite	1.7	2.8
Dicalcium phosphate	0.0	0.42
Calcium carbonate	0.27	0.10
Sodium chloride	0.50	0.50
L-Lysine HCl	0.064	0.0
DL-Methionine	0.114	0.104
L-Threonine	0.106	0.029
Vitamin/mineral premix	0.50	0.50
Robenidine premix	0.10	0.10

Table 2. Chemical composition of experimental diets (% DM)

<i>Chemical analysis</i>	Diets	
	1 & 3	2 & 4
Dry matter	91.3	91.5
Crude protein	17.2	16.9
Neutral detergent fibre	37.9	37.5
Acid detergent fibre	21.5	20.9
Acid detergent lignin	4.5	4.7

Table 3. Nutritive value

<i>Calculated values</i>	Diets
Digestible Energy (MJ/kg DM)	11.4
Digestible Protein (% DM)	12.7

Procedure

Forty-two New Zealand x Californian rabbits of 25 days of age, weighing 513 ± 66 (standard error, SE) g, were blocked by litter and randomly assigned to the experimental diets. No control of sex was done. Initial number of animals was 12 and 9 for treatments without or with antibiotic addition, respectively. During the trial seven animals in treatment 1 died because of enteritis. No losses occurred in any of the other treatments. Weight gain and feed intake were registered throughout a ten-days feeding period. After that, animals were slaughtered by cervical dislocation and a sample of 5 cm was taken from the jejunum.

Animals were individually caged and housed in a building in which the temperature was partially controlled (average room temperature $19.2 \pm 1.2^{\circ}\text{C}$). Animals were handled according to the principles for the care of animals in experimentation published by the Spanish Royal Decree 223/88. A cycle of 12 h of light and 12 h of dark was used throughout this trial.

Analytical methods

Jejunal samples were rinsed with 0.4 M KCl and were placed into a 10% buffered neutral formaldehyde solution (pH 7.2-7.4). All samples were gradually dehydrated through an increasing concentration of ethyl alcohol (50-100%). These specimens were first embedden in paraffin, and samples were prepared by sectioning at 6 μm and staining with hematoxylin and eosin. The intestinal histology was measured according to Hampson (1986). Both villus height and crypt depth were determined by averaging 30 independent measurements.

Diet dry matter (DM) was determined on duplicate samples by heating at 103°C for 24 h. Acid detergent fibre (ADF), acid detergent lignin (ADL) and neutral detergent fibre (NDF) were determined according to the sequential procedure of Van Soest et al, 1991. Procedure of the Association of Official Analytical Chemists (1995) was used for crude protein (N Kjeldhal and autoevaluation distillation unit Kilab nitrolab-auto: 954.01).

Statistical analysis

Data were analysed as a completely block design with litter as block effect. Main factors studied were inclusion of plasma, inclusion of antibiotics and its interaction. The General Linear Model (GLM) procedure of the Statistical Analysis Systems Institute Inc (SAS, 1990) was used. A t-test was used for means comparisons. Data are presented in tables as least-square means.

RESULTS AND DISCUSSION

Since the interaction plasma x antibiotic was significant, the combination of the experimental treatment are reported in table 4. Type of diet did not affect either feed intake or average daily gain during the experimental period (from 25 to 35 days of age), although they tended to be greater for the diet including both animal plasma and antibiotics (Table 4). Diarrhoea mortality was very high (54.5%) in the diet without animal plasma or antibiotic addition. No losses occurred in animals receiving any of the other diets.

Inclusion of animal plasma in the diet increased villus height and crypt depth ($P = 0.08$) by 31.2 and 14.4%, when no antibiotics were added to the diet. However, no effects of plasma addition on jejunal histology were found when antibiotics were present (Table 4). The low significance observed for the differences between treatments could be due to the small number of animals remaining in treatment 1 at the end of the trial.

These results agree with previous work in piglets (Touchette et al., 1997). They indicate that inclusion of animal plasma in starter diets for early-weaned rabbits might improve intestinal morphology when no antibiotics are added to the diet. The results obtained would contribute to explain the increases of intake and growth rate observed in similar conditions in another trial, where a larger number of animals was used (Gutiérrez et al., 2000).

Villus height and crypt depth obtained in this study were respectively shorter and greater than those obtained (Gutiérrez et al., unpublished) for 35-d rabbits that had assumed mother's milk until this age: $532 \mu\text{m} \pm 71.6$ (SE) and $68.5 \mu\text{m} \pm 6.45$ (SE). An intestinal atrophy after weaning has been also observed in early-weaned piglets by Allee and Touchette (1999), who related it to a lower digestion capability. Our results indicate that digestive and growth traits of early-weaned rabbits might be enhanced using highly digestible nutrients in the starter diet.

Table 4. Effect of supplementation with animal plasma and antibiotics on growth traits and the intestinal structure of early weaned rabbits (LS means \pm SEM¹)

	Diets			
	1	2	3	4
% animal plasma	0	4	0	4
Antibiotics	NO	NO	YES	YES
Rabbits, n = 35				
<i>Growth traits</i>				
Weight gain, g/d	30.3 \pm 5.63	29.0 \pm 3.48	30.6 \pm 3.98	31.8 \pm 4.20
Feed intake, g/d	67.9 \pm 7.83	65.9 \pm 4.84	67.3 \pm 5.53	69.6 \pm 5.83
<i>Jejunal histology (μm)</i>				
Villus height	342 ^a \pm 53.2	449 ^b \pm 29.1	418 ^b \pm 34.0	421 ^b \pm 34.0
Crypt depth	104 ^a \pm 7.07	119 ^b \pm 3.86	118 ^b \pm 4.52	110 ^{ab} \pm 4.52
Villus height/crypt depth	3.28 \pm 0.531	3.81 \pm 0.290	3.56 \pm 0.339	3.88 \pm 0.339

^{a,b} Means in the same row with different superscripts differ ($P < 0.10$)

¹ SEM: Standard Error of Means

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