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## GARCÍA J., NICODEMUS N., ESPINOSA A., CARABAÑO R., DE BLAS J.C.

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## CHARACTERIZATION OF FIBRE DIGESTION OF GRAPE-SEED MEAL AND SUNFLOWER HULLS IN RABBITS. II. CAECAL AND CAECOTROPHY TRAITS<sup>1</sup>

GARCÍA J., NICODEMUS N., ESPINOSA A., CARABAÑO R., DE BLAS J.C.

Departamento de Producción Animal, Universidad Politécnica de Madrid. ETS Ingenieros Agrónomos, Ciudad Universitaria. 28040 Madrid, Spain. jgarciaalonso@pan.etsia.upm.es

#### ABSTRACT

Twenty New Zealand White x Californian rabbits were used to evaluate excretion and composition of soft faeces and caecal traits of defatted grape-seed meal (GSM) and sunflower hulls (SH). Two diets were formulated by supplementing these fibrous feeds with a concentrate containing starch, protein, fat and a min/vit mix. Total and microbial nitrogen concentration were higher for animals fed SH than for those of GSM diet, but the higher soft faeces excretion obtained for GSM diet respect to SH avoid any difference in total and microbial nitrogen recycled through caecotrophy (0.58 and 0.26 g/d, on average). Either no differences were found between diets for DM intake and caecal ammonia concentration and pH (152 g/d, 23.7 mmol/l and 6.26, on average). Weights of stomach and of stomach contents were higher for SH diet compared to GSM (1.08 and 3.33 vs 0.90 and 2.35% BW, respectively). The higher hydration capacity and the proportion of particles larger than 1.25 mm of SH would explain these differences. Sunflower hulls diet also showed a higher weight of caecal digesta and therefore of total digestive tract respect to GSM. By comparing these results with previous literature, we conclude that GSM and SH might substitute partially alfalfa hay if they are combined with more fermentable sources of fibre that optimize caecal fermentation.

#### **INTRODUCTION**

The inclusion of alternative sources of fibre in rabbit diets can influence significantly rate of passage, caecal fermentation and soft faeces excretion (Fraga et al., 1991; García et al., 1999; García et al., 2000<sub>a</sub>). In some cases, these effects limit their inclusion in rabbit diets. Highly digestible sources of fibre such as sugar beet-pulp increase caecal acidity but also weight of caecal contents, which reduces feed intake and impairs performance (García et al., 1993; Carabaño et al., 1997). Inclusion of fibrous feeds such as wheat straw, with large particle size, decreases weight of caecal contents and caecal acidity (De Blas et al., 1986). In a previous work (García et al., 2000<sub>b</sub>) has been determined that GSM and SH impairs fibre digestion efficiency without affect rate of passage compared with alfalfa hay.

The aim of this work was to study caecotrophy and caecal traits of GSM and SH using semisynthetic diets that contained these feeds as sole source of fibre.

#### **MATERIAL AND METHODS**

#### Diets

Two diets were formulated to contain GSM and SH as the sole source of fibre. To assure a minimum nutrient supply, fibre sources were supplemented with a concentrate to

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obtain diets containing at least 18% crude protein and 12% starch. Chemical composition of fibre sources is shown in Table 1. Ingredient and chemical composition and particle size distribution of diets are shown in Table 2.

	Grape seed meal (GSM)	Sunflower hulls (SH)	
Dry matter	90.0	92.8	
Ash	5.84	3.01	
Ether extract	3.60	2.65	
Crude protein	11.0	5.56	
NDF	80.6	75.7	
ADF	72.0	59.7	
ADL	59.0	21.8	
Acid detergent cutin (ADC)	46.0	10.4	
GE, MJ/kg DM	20.5	20.3	
Dry density, g DM/ml	0.345	0.170	
Hydration capacity, %	192	506	

Table 1. Chemical composition of the fibrous raw materials (% of DM)

#### **Caecotrophy trial**

A group of 20 New Zealand White x Californian rabbits (ten per diet), 46 days old and weighing  $1324\pm36$  (SE) g, were randomly allotted to the experimental diets without regard to gender. Animals were given ad libitum access to the feed. Following a 28-d period of adaptation to each diet (after hard faeces collection) a wooden collar (150 g and 25 cm diameter) was put on each animal to prevent the ingestion of soft faeces as described by García et al. (2000<sub>a</sub>). In this period animals weighed 2027±60 (SE) g. The soft faeces collected were stored at -20° C, freeze-dried and total and microbial nitrogen analyzed. Data of two rabbits (one of each treatment) were removed due to their low soft faeces excretion.

#### **Caecal trial**

Five days after the last caecotrophy control, the animals were slaughtered by cervical dislocation at 19.00 h, to avoid the caecotrophy period. They weighed  $2261\pm58$  (SE) g. The gastrointestinal tract was removed and weighed. The stomach and caecum were weighed separately with and without their contents and pH was measured in the caecal contents. Cecal ammonia concentration was determined according to García et al. (2000<sub>a</sub>).

### Housing

Animals were housed in metabolism wired cages measuring 405 x 510 x 320 mm high that allowed separation of faeces and urine. A cycle of 12 h of light and 12 h of dark was used throughout the experiment. The light was switched on at 7.30 h. Heating and forced ventilation systems allowed the building temperature to be maintained between 18 and 23°C throughout the experiment. Experimental procedures followed the principles for care of animals in experimentation (Spanish Royal Decree 223/88, 1988).

#### **Analytical Methods**

Chemical analysis of diets and faeces was performed using the method of Van Soest et al. (1991) for NDF and Goering and Van Soest (1970) for ADF, ADL and acid detergent

cutin (ADC). Neutral detergent fibre was determined directly, whereas ADF and ADL were extracted successively. Acid detergent cutin was determined after extracting ADF, ADL and permanganate lignin. Procedures of AOAC (1990) were used for DM, ash, CP and ether extract. Proportions of fine and large particles (FP: < .315 mm, LP: > 1.25 mm, respectively) of diets were determined by wet sieving according to García et al. (1999). Dry density and hydration capacity were determined as described by Mir et al. (1990). Europium contents of marked fibre and faeces were analyzed by atomic emission spectrometry (Smith-Hieftje 22, Thermo Jarrell Ash, MA, USA) using predosed samples of faeces to prepare common-matrix standards. Previously, samples were ashed (550°C) and then digested by boiling with a solution of 1.5 M HNO3 and KCl (3.81 g/l).

	61.3% GSM	61.3% SH
Ingredient, % as fed		
Grape seed meal	61.3	0
Sunflower hulls	0	61.3
Pork lard	1.20	1.20
Wheat flour	18.0	18.0
Casein	15.3	15.3
Sepiolite	1.53	1.53
Calcium carbonate	.50	.50
Calcium phosphate	1.50	1.50
Sodium chloride	.50	.50
Vitamin/mineral premix <sup>a</sup>	.17	.17
Chemical analysis, % DM basis		
GE, MJ/kg DM	19.7	19.5
Ash	7.60	5.86
Crude protein	24.0	19.4
NDF	49.1	47.3
ADF	44.5	36.9
ADL	36.0	13.7
Acid detergent cutin	27.7	5.89
Particle size distribution, % DM		
< .160 mm	57.7	44.8
.160315 mm	13.6	11.1
.315630 mm	19.7	18.5
.630 – 1.250 mm	7.90	20.1
> 1.250 mm	1.10	5.5

Table 2. Ingredient and chemical composition of experimental diets.

<sup>a</sup> Provided by Roche Vitaminas (Madrid, Spain). Mineral and vitamin composition (mg/kg of feed): Mn, 22.8 (MnSO<sub>4</sub>); Zn, 68 (ZnSO<sub>4</sub>); I, 1.14 (KI); Fe, 40.8 (FeSO<sub>4</sub>); Cu, 13.6 (CuSO<sub>4</sub>); Co, .59 (CoSO<sub>4</sub>); Se, .14 (Na<sub>2</sub>SeO<sub>3</sub>); riboflavin, 3.6; calcium d-pantothenate, 12.4; nicotinic acid, 46.9; menadione sodium bisulfite, 1.4; vitamin E, 37.4; thiamine, 1.14; pyridoxine, 1.4; biotine, .08; folic acid, .8; vitamin B<sub>12</sub>, .014; vitamin A, 11,390 IU/kg and vitamin D<sub>3</sub>, 1,598 IU/kg.

#### **Statistical Analysis**

Data were analyzed by using the GLM procedure of SAS (1990) with type of diet as main factor.

## **RESULTS AND DISCUSSION**

Feed intake did not differ between GSM and SH (156 g DM/d on average, Table 3) as was observed previously by García et al. (2000<sub>b</sub>). Soft faeces excretion tended to be a 26% higher (P = 0.053) for animals fed GSM than for those fed SH, when soft faeces excretion was expressed as percentage of BW and day or 18%, when it was expressed as g/d. On the opposite, microbial nitrogen concentration in soft faeces decreased 18% in animals fed GSM respect those fed SH (P = 0.023), and total nitrogen also tended to be a 4% lower for animals fed GSM (P = 0.071). According to García et al. (2000<sub>a</sub>), these values are positively influenced by fine and large particle proportions and pectin constituents content and negatively by degree of lignification of NDF. In this case, the lower value of animals fed GSM respect to those of SH would be related with its higher degree of lignification and the presence of tannins, which would interfere with microbial activity, and possibly with a lower pectic constituents concentration.

As a consequence of the opposite effects of type of fibre on soft faeces excretion and its total and microbial nitrogen concentration, no difference were detected in the total and microbial nitrogen recycled daily through caecotrophy (0.58 and 0.26 g/d, on average, respectively). These values are lower than those found previously for alfalfa hay (1.0 and 0.66 g/d) or soya bean hulls (1.07 and 0.48 g/d), but very close to that obtained for NaOH-treated straw (0.57 and 0.34 g/d) (García et al., 2000<sub>a</sub>).

Table 3. Effect of inclusion of grape seed meal and sunflower hulls on excretio	n and	chemical
composition of soft faeces.		

	61.3% GSM	61.3% SH	<b>SEM</b> <sup>a</sup>	Р
DM intake the 3 previous days, g/d	154	158	6.79	0.67
Soft faeces excretion				
g DM/d	23.9	20.2	1.86	0.17
g DM/(100 g BW) d	1.21	0.96	0.08	0.053
Chemical composition, % DM basis				
Total nitrogen	2.59	2.70	0.04	0.071
Microbial nitrogen	1.08	1.32	0.068	0.023
Total nitrogen recycled daily through caecotrophy, g/d	0.62	0.55	0.046	0.30
Microbial nitrogen recycled daily through caecotrophy, g/d	0.26	0.26	0.027	0.94

 $^{a} n = 9$ 

There were no differences in caecal ammonia concentration and pH between animals fed GSM or SH, obtaining for both diets very high values (23.8 mmol/l and 6.26, on average, respectively) (Table 4) and similar to those observed previously for NaOH-treated straw (18.0 mmol/l and 6.28) by García et al. (2000<sub>a</sub>). This high concentration of ammonia in the caecum would be related with the low dietary digestible energy/digestible protein ratio of animals fed GSM and SH (46.9 and 56.4 Kj DE/g DCP, respectively, García et al, 2000<sub>b</sub>) (Fraga, 1998). This result indicates that the limiting factor on the synthesis of microbial nitrogen was not caecal ammonia concentration. In this sense, it seems that the reduced support of energy to caecal microorganisms, as NDF digestibility of this fibrous feeds is very low (García et al., 2000<sub>b</sub>), would be the factor that limits the caecal microbial activity. Accordingly, it would be

expected a low caecal volatile fatty acid concentration (not measured) and acidity, as it was observed.

Animals fed GSM had a weight of caecal contents 27% lighter than those fed SH (P = 0.001) which do not agree with their proportion of particles lower than 0.315 mm. So, it could have been expected a lower total and caecal mean retention time for GSM than for SH. However, García et al. (2000<sub>b</sub>) did not found differences in this variables between GSM and SH. These results could be explained by the higher soft faeces excretion of GSM respect to SH. This would mean that caecal contents of rabbits fed GSM are used in a major proportion to constitute soft faeces than those of SH, and thus total and caecal MRT did not differ respect to that of SH, although the weight of caecal content of animals fed GSM was lower.

Weight of caecum differ widely between rabbits fed these sources of fibre and was not parallel to that of caecal digesta. Weight of stomach and of stomach contents increased 20 and 40% from animals fed GSM with respect to those fed SH. This effect would be explained by the higher hydration capacity and proportion of particles larger than 1.25 mm of SH (García et al. 2000<sub>a</sub>). These variables are related with the bulk effect of the feed and would favour both a greater weight of stomach and stomach content and a longer mean retention time of digesta in the stomach. Total weight of digestive tract was higher for animals fed SH compared to those fed GSM in accordance with their higher weight of stomach and stomach and caecal digesta.

From these results it could be concluded that inclusion of GSM and SH in the diet impairs microbial nitrogen recycled through caecotrophy and increases caecal pH with respect to alfalfa hay. So it would be advisable to combine them with more fermentable sources of fibre in order to optimize caecal fermentation and avoid and excesive basicity of caecal digesta.

	61.3% GSM	61.3% SH	<b>SEM</b> <sup>a</sup>	Р
Dry matter intake, g/d	151	153	6.25	0.78
Digestive tract weight, %BW	16.1	17.8	0.46	0.014
Stomach weight, %BW	0.90	1.08	0.022	0.001
Stomach content weight, %BW	2.35	3.33	0.23	0.009
Caecal weight, %BW	2.01	1.60	0.067	0.001
Caecal content weight, %BW	3.63	4.96	0.17	0.001
Caecal pH	6.26	6.25	0.041	0.85
N-NH <sub>3</sub> , mmol/L	23.9	23.6	1.98	0.89

Table 4. Effect of inclusion of grape seed meal and sunflower hulls on caecal traits.

 $^{a} n = 10$ 

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