

SHORT NOTE [NOTA CORTA]

*Tropical and
Subtropical
Agroecosystems*

INFLUENCE OF BETAINE ON GOAT MILK YIELD AND BLOOD METABOLITES

[INFLUENCIA DE LA BETAINA SOBRE EL RENDIMIENTO LECHERO EN CABRAS Y SUS METABOLITOS EN SANGRE]

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SUMMARY

Betaine is a natural occurring compound with methyl donor properties which is increasingly being used in animal feeding. Betaine, an oxidative product of choline is able to replace methionine in some physiologically important body processes. The subject of this work was to study the effect of betaine added to the diet on milk production and blood metabolites on Murciano-Granadina dairy goats. Sixty lactating goats were selected from a commercial Murciano-Granadina goat herd (EXCAMUR S.L.) located in Murcia Region (Spain). Goats were selected from a 250 goats herd, taken into account the age, stage of lactation (2.5 as average), live weight (36 kg as average) and type of birth (2 kids). Two homogenous groups of 30 goats were made and fed with 1.5 kg of compound feed and 1 kg of alfalfa hay per day and goat. Goats were fed twice a day and water was provided *ad libitum*. Both groups received the same diet but for the second group the diet was supplemented with 4 g·kg⁻¹ betaine (betaine anhydrous, Danisco Animal Nutrition). The feeds, presented in pellets, were formulated in based on recommendations of INRA (2007). The experimental period was 6 months and the experimental diets were provided 15 days before parturition. The herd was machine milked once at day. Chemical composition, milk production and blood metabolites of each goat were recorded and analyzed at the end of the trial. Variance analysis and means comparison were carried out using the general lineal model procedure and Tukey test for mean comparison. Goats fed with betaine diet had higher milk fat than goats fed control diet (4.8 vs. 5.2 % for control and betaine respectively; P<0.05). No significant differences were found for milk production (1.8 kg d⁻¹ as average). Within blood metabolites, significant differences were found for the level of triglycerides, goats feeding with betaine were lower triglycerides level (0.76 vs. 0.48 mmol/l for control and betaine respectively; P<0.05). Further studies are necessary to understand the

mechanism of action of betaine on physical-chemical characteristics of milk.

Key word: goat, betaine, blood metabolites

INTRODUCTION

Heat is a major constraint on animal productivity in the tropical belt and arid areas. Mediterranean zones are exposed annually for 3-5 months of considerable heat stress. Exposure of goats to a high ambient temperature result in reduction of feed intake (Silanikove, 2000). Goats can mobilize their body reserves according to the climate conditions, nutritional status, physiological stage and availability of adipose tissue.

Betaine is a by-product of the sugar beet processing, and chemically is trimethyl derivat of the amino acid glycine. It is commercially available as a feed additive for livestock. Betaine is used by numerous tissues as an osmolite. The osmolytic activity of betaine has to be attributed to its dipolar zwitterions characteristics and its high solubility in water. Furthermore, betaine provides three methyl groups which can use in transmethylation reactions for the synthesis of numerous substances such as carnitine, creatine, phosphatidilcoline as well as methylated aminoacids. Due to both its methyl donor and its aminoacid function, betaine is involved in protein and energy metabolism (Eklund et al. 2005). Methyl groups are needed in the nervous, immune, renal and cardiovascular system. Moreover, under a stress situation or disease, methylation reactions are needed for building the immune defence mechanism as well the synthesis of polyamides, which play a role in tissue repair process (Virtanen and Rumsey, 1996). In the liver, betaine is a potent protector against bile acid induced apoptosis and, the major involvement of betaine in lipids metabolism is in its lipotropic activity (Eklund et al. 2005).

There is a lack of information of the effect of dietary betaine during lactation in dairy goats under harsh environment. The subject of this work

was to study the effect of betaine added to the diet on milk performance, including biochemical and hematologic parameters, on Murciano-Granadina dairy goats. Experiment will be conducted during the summer time in Murcia Region, characterised by high temperatures.

MATERIAL AND METHODS

Animal and diets

Sixty lactating goats were selected from an experimental Murciano-Granadina goat herd (EXCAMUR, S.L.) located in Murcia Region (Spain), which developed a good management practices. Goats were selected from a 250 goats herd, and age, stage of lactation (2.5 as average) type of birth (2 kids) and previous milk yield were taken into account. Two homogenous groups of 30 goats were made and they were fed with 1.5 kg of compound feed and 1 kg of alfalfa hay per day and goat. Goats were fed twice a day; 9:00 (after milking) and 15:00 with a half quantity each time and water was free available all the time. One group was fed the control diet, and the other the same diet containing 4 g·kg⁻¹ betaine (betaine anhydrous, Danisco Animal Nutrition). Betaine, raw materials and vitamin-mineral mix were provided by Unión Agropecuaria del Guadalentín S.A. and the diets

were pelleted. Balance of diets was obtained using the recommended values of INRA (2007). Ingredients and chemical composition of diets are shown in Table 1.

Experimental Procedure

All goats were housed in a building in which the environment (temperature) was partially controlled by two nearest Temperature/Humidity devices (HOB0® Data Loggers/Controllers) recording the daily maximum and minimum temperature. The average maximum and minimum of three days before milk control was used.

The experimental period took a period of time of 6 months and the intake of the experimental diets started 15 days before parturition. BCS was determined at the beginning and the end of the trial. The herd was machine milked once at day (morning milking) using a vacuum level of 40 Kpa, a pulsation frequency of 90 ppm and 60/40 as a relation of pulsation. Milk production and chemical composition of each goat was recorded and analyzed at the end of the experimental trial as blood samples. Body weight was recorded with an electronic scale (Gruponor-Cercampo) at the beginning and the end of the trial (36 ± 1.5 kg as average).

Table 1. Ingredients and chemical composition of diets.

Ingredients, g kg ⁻¹	Control	Betaine
	Corn grain	250
Barley grain	190	190
Rye grain	110	110
Carob bean	85	81
Beet liquid molasses	20	20
Alfalfa	30	30
Soybean meal, 44%	264	264
Fat	10	10
Betaine	-	4
Salt	7	7
Dicalcium Phosphate	14	14
Calcium carbonate	14	14
Vitamin /mineral mix ^a	5	5
Chemical analysis, %DM		
Dry Matter	88.7	88.5
Crude protein	15	15
NDF	24	24
Ether Extract	2.9	2.9

^a (ppm or UI per kilogram of diet) : Se, 40 ; I, 250 ; Co, 80 ; Cu, 3000 ; Fe, 6000 ; Zn, 23400 ; Mn, 29000 ; S, 60000 ; Mg, 60000 ; vitamin A, 2000000 UI ; vitamin D3, 400000 ; vitamin E, 2000 ppm ; nicotinic acid, 10000; choline, 20300.

Lactation experimental procedures were approved by the Committee on Animal Use and Care at the Universidad Politécnica de Valencia (Spain), and follow the codes of practice for animals used in experimental Works proposed by the European guidelines (European Union Directive No. 86/609/CEE, 2005).

Chemical Analyses of Diets and Milk

Chemical analyses of diets were carried out according to the methods of AOAC (1995). The dry matter (DM) of the diets was determined by oven-drying at $102 \pm 2^\circ\text{C}$ for 24 h, and the crude protein (CP) in a Kjeltec 2300 analyzer unit (Fosstecator). Ether extract (EE) was measured after acid hydrolysis to recover saponified fat with a DET-GRAS Selecta equipment, by extraction with diethyl ether. For neutral detergent fiber (NDF) the method of Robertson and Van Soest (1981) was followed and was determined by a filter bag technique (ANKOM Technology Corp. Fairport, NY, USA).

Physical-chemical parameters of the milk (Fat, crude protein and lactose) were determined by near infrared reflectance spectroscopy (NIRS, InfraAlyzer 500 D, Bran+Luebbe, Germany). SCC was determined in each goat at the end of the trial with a Fluoro-opto-electronic counter (Fossomatic 90, Foss Electric, Hillerød, Denmark). Physical-chemical analysis of diet and milk were determined at the Animal Science Lab from Universidad Politécnica de Valencia.

Blood Collection and Serum Chemistry Analysis

Whole blood was collected via jugular venipuncture into tubes containing ethylenediaminetetraacetic acid (EDTA) and serum separator Vacutainer tubes. Whole blood samples were gently mixed. Blood in serum separator tubes was allowed to clot for at least

30 min, and the tubes centrifuged within one hour to separate serum from the cells. Blood samples were kept cool and transported to laboratory (Laboratorio de Diagnostico General, Barcelona, Spain) for processing within 24 hr. Hematologic and biochemical analyses were completed within 48 hr.

Data Analysis

Data obtained after 6 months of lactation were analyzed by the software SAS (2001). Variance analysis and means comparison were carried out using the GLM procedure and Tukey test respectively.

RESULTS AND DISCUSSION

Average temperatures recorded during the experimental trial were 35°C as maxima and 22°C as minima. The BCS of the two groups of goats at the end of the trial (6 months) was 2 (poor BCS on a 1-5 scale).

Effect of betaine on milk performance was shown in Table 2. No significant differences were found for milk production (1.8 kg d^{-1} as average). In a study of Fernández et al. (2004) in primiparous lactating dairy goats using the same level of betine that in the present trial an increase of 0.28 kg d^{-1} in milk production was observed ($P < 0.05$) in goats fed betaine diet. In other previous study, Fernández et al. (2000) incorporated betaine into the diet at the level of 2 g kg^{-1} and a tendency ($P < 0.0947$) for milk yield was found in Murciano-Granadina dairy goats at second lactation for the group of goats feeding with betaine. Other experiment (Sánchez et al., 2001) using betaine at the level of 4 g kg^{-1} in primiparous goats after 4 months of lactation observed the same effect (increases of 0.21 kg d^{-1} for goat supplemented with betaine).

Table 2. Effect of dietary betaine administration on milk production and chemical composition in Murciano-granadina dairy goats.

	Control	Betaine	S.E.M.	<i>P</i>
Milk yield (ml/d)	1754.17	1996.67	576.858	NS
Fat (%)	4.8	5.2	0.155	0.04
Crude protein (%)	3.55	3.51	0.010	NS
Lactose (%)	4.71	4.73	0.009	NS
Log SSC	5.5	5.4	0.115	NS

We observed that goats fed with betaine diet had higher milk fat than goats fed control diet (4.8 vs. 5.2 % for control and betaine respectively; $P < 0.05$). In the study of Fernández et al. (2004) significant differences were found for milk fat (4.7 vs. 4.4% for betaine versus control respectively). Fernández et al. (2000) and Sánchez et al. (2001) did not found significant differences for chemical composition of milk; the values of fat were 4.93 and 4.84% for betaine and control diet, respectively. We must indicate that the present trial was run during the summer time and the trials of Fernández et al. (2000; 2004) and Sánchez et al. (2001) was conducted during spring. No significant differences were found for milk protein, lactose and log SCC.

Blood metabolites are shown in Table 3. Within blood metabolites, significant differences were found for the level of triglycerides, so goats feeding with betaine were lower triglycerides level than the control group (0.76 vs. 0.48 mmol/l for control and betaine respectively; $P < 0.05$). Lower ketone bodies ($P < 0.05$) were also found for betaine vs. control group (0.383 vs. 0.347 mg/dl for control and betaine respectively). Caldeira et al. (2007a) found higher levels of serum triglycerides at extremes BCS, and can be explained by the abundance of precursors of alimentary origin at the high BCS, and of non esterified fatty acid from fat mobilization at lower BCS. Regards ketone bodies, the same authors indicated the first decline of β -hydroxybutirate concentration at a BCS of 3.5 (following the implementation of feed restriction) and then

increased at a BCS from 3.5 to 2.5, as a partial oxidation of the mobilized non esterified fatty acid to β -hydroxybutirate from body reserves. Although we did not found significant differences for the rest of blood metabolites, Caldeira et al. (2007b) observed that undernutrition with a BCS of 2 and, particularly 1.25 in ewes was illustrated by low concentration of plasma glucose, albumin, globulins and high concentration of urea and creatinine.

Mitchell et al. (1979) reported approximately 50% of the carboxyl and methyl carbons of betaine were presumably retained within the animal 10 to 14 hrs following the addition of betaine to the rumen. Even though betaine disappeared from the rumen quite rapidly it is conceivable that some of the betaine escaped rumen metabolism to be absorbed and participate in the methyl-transfer reactions in the liver. Although we did not find information about the effect of betaine added to the diet under high temperatures in dairy goats, seems that betaine has a positive effect on lipid metabolism.

CONCLUSIONS

Oral supplementation with betaine (4 g kg⁻¹) during summer time increased the level of milk fat at late lactation. Lower levels of triglycerides and ketone bodies were observed in the fed betaine group. Further studies are necessary to understand the mechanism of action of betaine on metabolism under heat stress conditions.

Table 3. Effect of dietary betaine administration on serum metabolites in Murciano-Granadina dairy goats.

	Control	Betaine	S.E.M.	<i>P</i>
Glucose (mmol/l)	3.056	3.294	0.556	NS
Urea N (mmol/l)	7.706	7.225	2.050	NS
Triglycerides (mmol/l)	0.76	0.48	0.28	0.011
Creatinine (μ mol/l)	67.92	70.50	7.072	NS
Total protein (g/l)	79.96	78.61	5.428	NS
Albumin (g/l)	43.450	41.558	3.3935	NS
Ketone bodies (mg/dl)	0.383	0.347	0.1189	0.046

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