Nutritional Utilization in Malagueña Dairy Goats Differing in Genotypes for the Content of α_{S1} -Casein in Milk

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ABSTRACT

A study was carried out with 20 goats of the Malagueña breed, half with a high (HG) and half with a low (LG) genetic capability for α_{S1} -casein (AS1-CN) synthesis, to determine whether the 2 different genotypes (that cause differences in goat milk composition) are related to differences in nutritional feed utilization. Among the 10 HG goats, 7 had BB and 3 had AB genotypes for *AS1-CN*, whereas there were 7 EF and 3 FF genotypes in the 10 LG goats. The goats were fed diets differing in crude protein content (13.6 vs. 17.7% dry matter for diets 1 and 2, respectively). For each genotype group, a balance trial was conducted with each of the 2 diets in a 2-period balanced changeover designed with half the animals consuming diet 1 and the other half diet 2, determining individual feed intake and the utilization of N and energy in the diets. Greater voluntary feed intake on a metabolic body weight basis among the HG goats was identified as the first possible cause of their milk production. The HG goats also had a greater level of feed utilization, on a metabolic body weight basis, for N and energy intake. Greater ratios of N balance/digestible N, milk protein N/digestible N, milk energy/digestible energy, and milk energy/ME were found for HG goats compared with LG. These effects appear to be dependent on the level of protein in the diet, indicating interactive effects. The greater N and energy utilization of HG versus LG goats may explain the differences in milk composition between the 2 genotype groups.

Key words: intake level, feed N and energy utilization, genotype for α_{S1} -casein, lactating goat

INTRODUCTION

In goats, the gene for α_{S1} -casein (*AS1-CN*) in milk has an unusually high degree of genetic polymorphism with 18 allele types (Sacchi et al., 2005) that have been stud-

ied intensively since the 1980s (Boulanger et al., 1984). Grosclaude et al. (1987) analyzed the effects of the different alleles on the level of AS1-CN synthesis in Alpine and Saanen goats, and classified the 7 alleles into 4 groups: alleles A, B, and C with a high level of milk protein synthesis (3.6 g/L), allele E with an intermediate level (1.6 g/L), allele F with a low level (0.6 g/L), and the null allele with 0.0 g/L. This same polymorphism has been observed in other European goat breeds as well as in Spanish goat breeds (Serradilla, 2002). The milk produced by goats with a high capacity for the synthesis of AS1-CN also tends to have greater concentrations of milk fat (Barbieri et al., 1995).

Among factors of nutritional feed utilization that determine the productive capacity of goats it has been reported that those with a high capacity for the synthesis of AS1-CN also have greater feed intake per metabolic BW (**MBW**) than those with a low capacity (Schmidely et al., 2002; De la Torre, 2006). Increased levels of energy intake, depending on the genetic merit of the animal, may lead to greater production of milk or greater concentration of protein and fat in the milk (Morand-Fehr et al., 2000; Boza, 2005). However, the differences in voluntary feed intake (**VFI**) do not seem to justify the differences found for the amount of protein, casein, and fat in the milk. Therefore, in addition to different intakes, there could be different nutritional utilization. Possible variations in protein requirements depending on the genotype associated with AS1-CN level in milk have still to be clarified. This paper describes results from a study with Malagueña dairy goats belonging to genotypes for either low or high capacity to synthesize AS1-CN in milk. The goats were fed 2 diets varying in protein contents, and the utilization of N and energy was determined for individual goats. The study of the utilization of N and energy in the diet was also undertaken to determine possible genetic $\times$ nutrition interactions in relation to genotype and protein content of the feed.

MATERIALS AND METHODS

Experimental Design and Procedure

Twenty Malagueña goats, from 4 herds, midway through their second, third, or fourth lactation were

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Table 1. Ingredient composition of the concentrates, chemical composition, and gross energy content of the concentrates, alfalfa hay, and barley straw

Item	Concentrate ¹		Alfalfa hay	Barley straw
	D1	D2		
Ingredient, g/kg				
Oats	293	150		
Corn	390	75		
Faba beans	240	335		
Bypass fat	37	—		
Corn gluten feed	—	200		
Cottonseed	—	200		
Mineral-vitamin supplement ²	40	40		
Chemical composition and gross energy content	Concentrate ¹		Alfalfa hay	Barley straw
DM, %	89.1	89.5	91.8	89.4
			% of DM	
OM	92.1	91.7	92.5	94.8
CP	12.7	19.6	18.6	4.7
Fat	6.8	6.5	10.0	—
NDF	27.8	36.7	50.1	74.1
ADF	8.6	16.3	3.7	4.6
Acid detergent lignin	0.9	3.5	9.2	5.2
Gross energy, MJ/kg of DM	18.6	18.5	19.0	18.2

¹Concentrate: D1 = 13.6% CP (DM basis); D2 = 17.7% CP (DM basis).

²Mineral-vitamin supplement composition (% of DM): 5.8% Ca, 17.1% P, 25.0% NaCl, 2.3% Fe, 0.3% Cu, 1.5% Zn, 1.2% Mn, 3.0% Mg, 0.05% Co, 2.0% vitamin A, 0.13% vitamin D₃, 1.3% vitamin E, 0.8% nicotinic acid, 0.25% vitamin B₁, B₂, and B₁₂ (Musal Laboratory, Granada, Spain).

used. Ten of these animals had genotypes (7 BB and 3 AB) associated with a high level of α_{S1} -casein (AS1-CN) and total protein in milk (HG), and 10 had genotypes (7 EF and 3 FF) associated with a low level of AS1-CN and total protein in milk (LG). Each genotype group was made up of goats from all 4 herds to randomize the genetic backgrounds. Samples were taken from 300 animals for genotyping, which was performed following the protocol developed by Amills (1996). The animals were fed 2 diets differing in their CP content: 13.6% (diet 1, D1) and 17.7% (diet 2, D2) of DM. The proportion of degradable protein in the rumen corresponding to the diets was very similar ($74.6 \pm 0.6\%$). An in vivo balance trial was conducted with the HG goats, using the 2 different diets in a 2-period balanced changeover design. Within each period, half of the animals consumed D1 and the other half consumed D2. The same procedure was used with the LG goats. Each goat received, at the same time and in separate containers, a daily ration consisting of 1.0 kg of forage and 1.0 kg of concentrate (Aguilera et al., 1990; NRC, 2007). The forage fraction of the diets consisted of long alfalfa hay (700 and 800 g per animal and day for D1 and D2, respectively) and barley straw (300 and 200 g per animal and day for D1 and D2, respectively). Table 1 shows the ingredient composition of the concentrates and the chemical composition and energy content of the concentrates and alfalfa hay and barley straw used. The total duration of each

trial was 21 d, the first 14 d of which were for adaptation and the last 7 d constituting the main trial period. During the main trial period the goats were housed individually in metabolism cages. The goats were hand milked daily at 0900 h, once the orts from the previous day had been collected; daily rations were fed immediately thereafter. Water was available at all times. After milking, the goats were weighed on arrival at the laboratory and at the start and finish of the balance trials.

Measurements and Analysis

Daily samples of forage and concentrate together with samples of orts, feces, and urine were collected and maintained at -20°C until analysis. Similarly, samples of milk with no added preservatives were stored at -30°C until analysis. The DM and N contents were determined using fresh samples and all other analyses were carried out using dry samples. The DM content of the feedstuffs and orts were determined (AOAC, 2005), and that of the feces, urine, and milk were determined by lyophilization. The N content of the feedstuffs, orts, feces, urine, and milk were determined by the Kjeldahl method (AOAC, 2005). The protein N content of the milk samples was calculated as the difference between total N and NPN. Total N was determined from whole milk samples and NPN from a filtrate of whole milk after precipitation with 12% (wt/vol) TCA. Neutral detergent fiber, ADF,

Table 2. Effect of type of diet¹ and animal genotype² on BW, nitrogen intake, and excretion

Item	D1		D2		SE	Level of significance ³		
	LG	HG	LG	HG		Diet	Genotype	Diet $\times$ genotype
BW, kg	47.4	44.3	48.3	41.9	2.5	NS	**	NS
	— g of N/metabolic BW —							
Intake N	1.717	1.999	2.397	2.568	0.169	***	*	NS
Fecal N	0.490	0.554	0.653	0.708	0.061	***	*	NS
Urine N	0.610	0.648	0.987	1.021	0.079	***	NS	NS
Digestible N ⁴	1.227	1.445	1.744	1.860	0.117	***	*	NS
N balance ⁵	0.617	0.797	0.757	0.839	0.073	*	**	NS
Milk total N	0.336	0.419	0.420	0.428	0.032	*	*	*
Milk protein N	0.312	0.395	0.387	0.400	0.030	*	**	NS
N retained ⁶	0.281	0.378	0.337	0.411	0.065	*	*	NS
	— % —							
Digestible N/intake N	71.6	72.3	72.8	72.4	1.3	NS	NS	NS
N balance/digestible N	50.3	55.1	43.4	45.1	3.4	***	*	NS
Milk protein N/digestible N	25.4	27.3	22.2	21.5	3.1	*	NS	*
Milk protein N/N balance	50.6	49.6	51.1	47.7	6.1	NS	NS	NS
Milk protein N/milk total N	92.9	94.3	92.1	93.4	0.5	*	***	NS

¹Diet: D1 = 13.6% CP (DM basis); D2 = 17.7% CP (DM basis).

²Animal genotype: LG = group of goats with low capability for synthesis of α_{S1} -casein; HG = group of goats with high capability for synthesis of α_{S1} -casein.

³Diet = main effect of diet; genotype = main effect of animal genotype; diet $\times$ genotype = diet $\times$ animal genotype interaction.

⁴Digestible N = intake N – fecal N.

⁵N balance = digestible N – urine N.

⁶N retained = N balance – milk total N.

* $P \leq 0.05$; ** $P \leq 0.01$; *** $P \leq 0.001$; NS = $P > 0.05$.

and acid detergent lignin (ADL) contents of the feed-stuffs were determined (Van Soest et al., 1991), and fat content was determined after hydrochloric hydrolysis by extraction with petroleum ether (boiling point, 40 to 60°C; Sanderson, 1986). The ash content was determined by incineration in an electric muffle furnace at 550°C and the energy content of the different samples by adiabatic bomb calorimetry. Apparent N digestibility and the N and energy balances were calculated from the corresponding feed intakes, fecal and urinary excretion, and milk output. Methane energy output was estimated according to Aguilera et al. (1990) as 9.09% of digestible energy.

Statistical Analysis

Data were analyzed as a 2×2 factorial arrangement (Statgraphics, 2001); main effects were 2 diets and 2 genotypes in a completely randomized design. When the interaction term was not statistically significant ($P > 0.05$), the least squares means were calculated from the model after this term was omitted (Steel and Torrie, 1980). The dependent variables in the model were N and energy intake data as well as the N and energy balance data. Duncan's multiple range test ($P < 0.05$) was used to determine the differences between the means.

RESULTS

Table 2 shows the results obtained for utilization of N, by diet consumed and animal genotype, as well as the results of the statistical analysis. The dependent variables measured included N intake, fecal and urine N excretion, total N in milk and milk protein N, digestible N (N intake less fecal excretion), N balance (digested N less urine excretion), and N retained (N balance less total milk N). All values are expressed as grams per MBW per day. To analyze the efficiencies with which N intake was utilized for digestion, metabolism, and milk production, the ratios indicative of such efficiencies were calculated. Diet affected ($P < 0.05$) values of N intake, fecal N excretion, urine loss of N, digestible N, N balance, total N in milk, protein N in milk, N retained, N balance/digestible N, milk protein N/digestible N, and milk protein N/milk total N. Greater values were obtained for the goats in the D2 groups for N intake, fecal N excretion, urine loss of N, digestible N, N balance, total N in milk, protein N in milk, and N retained. However, lower values were obtained for the goats in the D2 groups, for N balance/digestible N, milk protein N/digestible N and milk protein N/milk total N. Genotype affected ($P < 0.05$) values of N intake, fecal N excretion, digestible N, N balance, total N in milk, protein N in milk, N retained,

Table 3. Effect of type of diet¹ and animal genotype² on energy intake and excretion

Item	D1		D2		SE	Level of significance ³		
	LG	HG	LG	HG		Diet	Genotype	Diet × genotype
	kJ/metabolic BW							
Gross energy	1,429	1,657	1,550	1,643	115	NS	*	NS
Fecal energy	410	480	463	480	34	NS	*	NS
Urine energy	31	33	50	51	4	***	NS	NS
Digestible energy ⁴	1,019	1,177	1,087	1,163	81	NS	*	NS
Methane energy ⁵	93	106	99	106	8	NS	*	NS
ME ⁶	895	1,038	938	1,006	71	NS	*	NS
Milk energy	208	293	260	264	26	NS	**	*
	%							
Digestible energy/gross energy	71.3	71.0	70.1	70.8	1.9	NS	NS	NS
Milk energy/digestible energy	20.4	24.9	23.9	22.7	2.2	NS	NS	*
Milk energy/ME	23.2	28.2	27.7	26.2	2.5	NS	NS	*

¹Diet: D1 = 13.6% CP (DM basis); D2 = 17.7% CP (DM basis).

²Animal genotype: LG = group of goats with low capability for synthesis of α_{S1} -casein; HG = group of goats with high capability for synthesis of α_{S1} -casein.

³Diet = main effect of diet; genotype = main effect of animal genotype; diet × genotype = diet × animal genotype interaction.

⁴Digestible energy = gross energy – fecal energy.

⁵Methane energy = methane output calculated from Aguilera et al. (1990) as 9.09% of digestible energy.

⁶ME = digestible energy – urine energy – methane energy.

* $P \leq 0.05$; ** $P \leq 0.01$; *** $P \leq 0.001$; NS = $P > 0.05$.

N balance/digestible N, and milk protein N/milk total N. All these values were greater for the HG goats. The interaction between factors for N balance ($P = 0.10$), milk total N ($P = 0.05$), milk protein N ($P = 0.07$) and N balance/digestible N ($P = 0.10$) reveals that the effect of genotype was in these cases, more apparent when D1 was consumed. However, the interaction between factors for milk protein N/digestible N ($P = 0.05$), indicates that the values for the HG goats were greater than those for the LG goats when D1 was consumed.

Total energy intake, fecal and urine energy excretion, losses as methane, the amount of digestible energy (DE, energy intake less fecal excretion), ME, and the energy in the milk (all expressed as kilojoules per MBW per day), and the energy efficiencies are given in Table 3. Diet affected ($P < 0.05$) urine energy losses, with greater losses for the goats in the D2 group. Genotype affected ($P < 0.05$) total energy intake, fecal losses, DE, CH₄ losses, ME, and energy in the milk, with greater values for the HG goats. The interactions between factors for energy in the milk, milk energy/DE, and milk energy/ME was significant ($P < 0.05$). The values for the HG goats were greater than those for LG goats when the D1 diet was consumed.

DISCUSSION

Utilization of Nitrogen

Our data (Table 2) were very similar to those of Lindberg and Gonda (1996) who reported that, in lactating

goats, the largest loss of the diet-derived intake of N is via urinary excretion (43%), followed by fecal excretion (29%) and the N in the milk produced (24%). In our study, values for LG goats given the D1 diet were 43, 34, and 23% for urinary N excretion, fecal N excretion, and N in the milk produced, respectively, whereas values in HG goats given the same diet were 40, 34, and 26%, respectively. Values for LG goats given the D2 diet were 48, 32, and 20% for urinary N excretion, fecal N excretion, and N in the milk produced respectively, whereas those in HG goats given the same diet were 47, 33, and 20% for urinary N excretion, fecal N excretion, and N in the milk produced respectively.

As N intake increases, fecal N, urine N, total N absorption, and milk N also increase (Badamana and Sutton, 1992; Kadzere and Jingura, 1993; Schmidely et al., 2002). Our data are similar. In agreement with Schmidely et al. (2002), we found that variations in the amounts of N retained in the body also increased in parallel with increasing levels of dietary protein; Badamana and Sutton (1992) reported different results. The utilization of protein or N in goats increases as the proportion of dietary protein increases, especially for diets with protein of low quality (Kadzere and Jingura, 1993; Swanson et al., 2000; Haddad et al., 2005). The efficiency with which the MP available is utilized for productive purposes is known to decrease as the levels of protein in the diet increase (Park, 1982). This has been confirmed in goats for growth (Sanz Sampelayo et al., 1995) and for milk production (Badamana and Sutton, 1992).

Table 4. Changes in analyzed values for goats with low (LG) or high (HG) capability for synthesis of α_{S1} -casein and fed D1 (13.6% CP of DM) or D2 (17.7% CP of DM)¹

Item	Difference between genetic groups ²		Difference between diets ³	
	D1	D2	LG	HG
	%			
N balance	+29.2	+10.8	+22.7	+5.3
Milk protein N	+26.6	+3.4	+24.0	+1.3
Milk total N	+24.7	+1.9	+25.0	+2.1
N balance/digestible N	+9.5	+3.9	-13.7	-18.1
Milk protein N/digestible N	+7.5	-3.2	-12.6	-21.2
Milk energy	+40.9	+1.5	+25.0	-9.9
Milk energy/digestible energy	+22.1	-5.0	+17.2	-8.8
Milk energy/ME	+21.6	-5.4	+19.4	-7.1

¹Values are presented for interactions between factors (diet $\times$ genotype) at $P \leq 0.10$.

²Values are reported as mean differences between genetic groups expressed as percentage [(HG - LG)/LG] $\times$ 100.

³Values are reported as mean differences between diets expressed as percentage [(D2 - D1)/D1] $\times$ 100.

In this study, the efficiencies of using digestible N for N balance or for milk N were lower when D2 was consumed.

The greater N intake by the HG goats, irrespective of the level of protein in the diet, was the consequence of greater feed intake (Schmidely et al., 2002; De la Torre, 2006). In goats, energy intake is the principal factor determining the protein content of the milk produced (Morand-Fehr et al., 2000; Boza, 2005). Therefore, the greater capacity for feed intake should determine the greater capacity for milk production. Second, there was greater fecal N excretion as well as greater quantities of digestible N, N balance, milk N, and N retained within the body. However, urinary N excretion was unaffected. Despite the different levels of intake, this lack of effect of the genotype on urinary N loss should be considered of special interest, because it suggests that the HG goats achieved greater efficiency in the utilization of digestible N for milk production. The milk produced by HG goats had a greater proportion of protein N with respect to total N, because of the lower quantity of NPN. The greater efficiency of HG goats in the utilization of MP for the production of milk protein depends on the protein content of the diet. This is an issue that requires more research. This is of vital practical interest, because it provides the foundation on which to modulate gene expression by means of nutrition.

The MP efficiencies for lactating goats reported herein are different than those reported for cows of diverse genetic types. McGilloway and Mayne (2002), studying cows of great genetic merit, reported that absorbed nutrients are apportioned more to milk production than to fat accretion. In the present study, although the HG goats were found to have lower average BW than the LG goats; the changes observed within each group between the start and end of the trials were less than ± 0.60 kg, which could simply be the result of differences in gut

fill. Furthermore, the milk protein N/N balance ratio was not affected ($P > 0.05$) by genotype.

The greater capacity for feed intake and the increased utilization of MP by HG goats compared with LG seems to explain why, under forced feeding conditions, the protein retention achieved is no greater than that resulting from ad libitum feeding conditions (McCrackens et al., 1994). The HG goats, in contrast to LG, possess a greater capacity for the utilization of MP, regulating their VFI in accordance with this greater capacity.

Utilization of Energy

It was not possible to determine ME intake in our study. When we consider a single class of animals consuming similar diets, the values of ME can be estimated on the basis of DE (Sanz Sampelayo et al., 2002). We found that MP was greater in HG goats, which suggests that the conversion of DE into ME must depend on the genotype. Metabolizable energy was estimated as being the difference between DE and the total energy losses as urine and methane. These latter values were estimated to be 9.09% of digestible energy (Aguilera et al., 1990). Gordon et al. (1995) reported that this loss does not vary between animals of diverse genotypes.

Despite the greater feed intake by HG goats, loss of energy via urine did not differ between genotypes within each diet. Goats with a high capacity for synthesis of α_{S1} -CN are more energy efficient. The energy converted into milk as well as the efficiency with which DE or ME was utilized for milk production were greater for HG goats given the D1 diet. The HG goats are capable of producing milk with greater protein and fat contents (Barbieri et al., 1995).

There is little information available on the utilization of energy in the lactating goat by different genotypes. In

dairy cattle of greater genetic merit, energy is apportioned in a greater proportion to the production of milk than to fat accretion (Gordon et al., 1995; Agnew et al., 2002); our results with Malagueña goats were similar.

Implications

The values consigned in Table 4 were derived from the values consigned in Tables 2 and 3. Compared with LG goats on the D1 diet, HG goats consuming D1 or LG goats consuming D2 had improved N balance, milk total N, milk protein N, milk energy, and greater efficiencies of utilization of DE or ME for milk production. The use of HG goats and changing from D1 to D2, or the use of the D2 diet and changing from LG to HG goats would be disadvantageous.

Greater VFI most affected the differences in the milk composition of dairy goats and the synthesis of AS1-CN. Malagueña goats also presented a greater degree of metabolic utilization, both of N and energy consumed. There appears to be a nutrition $\times$ genotype interaction with regards to AS1-CN synthesis.

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