Use of Different Dietary Protein Sources for Lactating Goats: Milk Production and Composition as Functions of Protein Degradability and Amino Acid Composition[1]

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ABSTRACT

To establish the effect of the nature of four different protein sources [fababeans, 27.8% crude protein (CP); sunflower meal, 41.7% CP; corn gluten feed, 18.8% CP; and cottonseed, 18.3% CP] on milk protein production by goats, the ruminal degradation of these feeds was studied as was the amino acid (AA) composition of the original material and that of the undegradable fractions of the protein sources. Four diets were designed; 20% of their protein was supplied by each of the different sources. Four groups of 5 Granadina goats were used to study the utilization of these diets for milk production. No significant differences were observed in dry matter intake or milk production. The milk produced by goats fed the diet containing sunflower meal had the lowest protein concentration; the highest milk protein concentration was observed for goats fed the diet containing corn gluten feed. From a multivariate analysis, it was deduced that the quickly degradable protein fraction in the rumen and the ruminally undegradable protein fraction were the components of the protein sources most directly related to the milk protein produced. Given the similar AA profiles of the undegradable fractions of the different protein sources, the possible supplementation achieved from these ruminally undegradable fractions must be established by the amount of protein supplied regardless of AA composition.

(**Key words**: protein source, degradability, milk production, lactating goats)

Abbreviation key: **CGF** = corn gluten feed, **CS** = cottonseed, **EAA** = essential AA, **FB** = fababeans, **SFM** = sunflower meal.

INTRODUCTION

The systems currently in use to estimate the protein requirements of ruminant animals distinguish between the dietary protein fraction, which after ruminal degradation yields microbial protein, and the fraction that escapes ruminal fermentation and reaches the small intestine (1, 20). Since the introduction of these new systems, there has been much research (5, 27) to determine the effect of protein degradability on milk production and composition. All of these studies have attempted to optimize milk protein production, but the results have not always been predictable. Different researchers (5, 15, 26, 27, 31) have pointed out the inadequacy of using only the intake of RDP and RUP to formulate diets for lactating ruminants. In their opinions, it would be necessary to take into account the kinetics of ruminal degradation of the protein supplied and the amino acid profile of the RUP fraction of the protein supplied.

The limited amount of research to date carried out in goats has only analyzed the effect of using less degradable protein sources instead of soya protein. Very few and even contradictory results have been obtained (11, 19). Morand-Fehr et al. (19) have stressed that in the majority of cases in which isonitrogenous and isoenergetic diets were used, the total protein and casein contents of goat milk did not appear to be very sensitive to changes in dietary protein source. Because of the scarce information available and because practically all of the goat milk produced in Spain is destined for cheese production, we analyzed the effect of using different protein

Received March 6, 1998.
Accepted September 14, 1998.
[1]This study was supported financially by the Interministerial Commision of Science and Technology, Madrid, Spain (Project AGF93-0096 and ALI96-1024-C02-01).

sources with different ruminal degradation characteristics: fababeans (**FB**), sunflower meal (**SFM**), corn gluten feed (**CGF**), and cottonseed (**CS**) on the production and composition of the milk of Granadina goats. The objectives of this study were to establish the overall changes in AA composition of the protein sources used that were due to ruminal incubation and to establish the pattern of relationship between those variables that defined the nature of the protein sources used and those variables that defined the amount and composition of the milk protein produced.

MATERIALS AND METHODS

Experimental Design

Two types of assays were performed. One of the assays analyzed the characteristics of the ruminal degradation of each protein source, examining at the same time the AA composition of each source before and after ruminal fermentation. The other set of assays used groups of Granadina goats to determine the effect on milk production and composition of diets in which part of the protein content was provided by the alternative protein sources.

Degradability and AA Composition of the Different Protein Sources

The ruminal degradability of the N in each protein supplement was estimated using the nylon bag technique according to an international standard procedure (16). Two wethers with permanent ruminal cannulas were used for incubations of 0, 4, 12, 24, 48, and 72 h. The diet fed to the cannulated wethers was that proposed by Ørskov and McDonald (21) and Isac et al. (13). The diet consisted of a good quality alfalfa hay with a mineral and vitamin supplement. For each incubation time, two bags were used per animal. The dimensions of the bags were 7.5×10 cm, and the pore size was 46×46 μm. The amount of sample per bag was 2 to 3 g, which were previously milled through a 2.0-mm screen. The 72-h incubation was repeated several times, and the residues were pooled to obtain sufficient material for AA analysis. After incubation, the bags were washed in a washing machine and were treated in a stomacher for 5 min to remove microbial contamination (16).

Milk Production and Composition

Twenty Granadina goats that were midway through their second lactation were divided into four equal groups based on BW and milk production. The goats (14) were selected to have the same protein polymorphisms. Before the experiment, the goats consumed the experimental diets for 1 mo. Then, the goats were housed individually in crates for 19 more d. Every goat received a daily ration consisting of 1.0 kg of alfalfa hay and 1.0 kg of concentrate; the specific N and energy requirements of this species and breed were considered in the dietary formulation (2). Treatments consisted of four diets, and 20% of the total protein was supplied by the four experimental proteins: FB, SFM, CGF, and CS. The four diets were similar in terms of N and gross energy contents. The ingredient composition of the concentrates and the chemical composition of diets are shown in Tables 1 and 2, respectively.

TABLE 1. Ingredient composition of the concentrates.

Ingredient	Diet[1]			
	FB	SFM	CGF	CS
	(g/kg)			
Oats	360	360	360	360
Corn	360	400	200	200
FB	240	...	...	...
SFM	...	200	...	...
CGF	...	...	400	...
CS	...	...	...	400
Mineral and vitamin supplement[2]	40	40	40	40

[1]Twenty percent of protein was provided by fababeans (FB), sunflower meal (SFM), corn gluten feed (CGF), and cottonseed (CS).

[2]Contained per kilogram: 2.32 g of Ca, 6.84 g of P, 10.0 g of NaCl, 0.92 g of Fe, 0.12 g of Cu, 0.60 g of Zn, 0.48 g of Mn, 1.20 g of Mg, 0.02 g of Co, 1.33 million IU of vitamin A, 2.08 million IU of vitamin D_3, 520 IU of vitamin E, 0.32 g of nicotinic acid, and 0.10 g of vitamins B_1, B_6, and B_{12}.

The first 15 d of the experimental period were for adaptation, and the last 4 d constituted the principal trial period. At 0900 h every day, once the orts from the ration that was offered the previous day had been collected, the goats were hand-milked. Subsequently, the daily rations were distributed. Water was available at all times.

After milking, the goats were weighed on the 1st, 15th, and 19th d of the experimental period. Feed intake and milk production were monitored daily.

Measurements and Analyses

Every day of the principal trial period, samples of the forage and concentrate offered and samples of orts were collected to determine the composition of the diet fed and consumed. Similarly, samples of milk

TABLE 2. Chemical composition and gross energy content of diets.

	Diet[1]			
	FB	SFM	CGF	CS
DM, %	88.40	88.49	88.73	89.39
	(% of DM)			
OM	90.26	89.78	88.76	90.02
CP	18.00	19.08	17.63	17.60
Fat	2.47	2.43	2.80	5.70
NDF	35.49	36.65	38.14	41.21
ADF	21.24	22.45	21.58	27.50
ADL[2]	4.39	4.96	4.15	7.55
Gross energy, MJ/kg of DM	18.1	18.1	17.9	18.9

[1]Twenty percent of protein was provided by fababeans (FB), sunflower meal (SFM), corn gluten feed (CGF), and cottonseed (CS).

[2]Acid detergent lignin.

with no added preservatives were stored at –30°C until analysis.

The DM and N contents of the dietary component samples and orts, milk, and milk fat were analyzed from fresh samples. All other analyses were performed on dried samples. The DM of the feedstuffs was determined by oven-drying at 100 ± 2°C for 24 h. The N contents were measured using the Kjeldahl method (3). These values were converted to CP by multiplying by a factor of 6.25. The NDF, ADF, and acid detergent lignin contents were determined using the method of Goering and Van Soest (10). The fat contents were measured by extraction with petroleum ether (boiling point, 40 to 60°C). The DM analysis of milk was carried out by lyophilization. The N content was measured using the Kjeldahl method (3). Protein nitrogen content of the milk samples was calculated as differences between total N and NPN; total N was determined from whole milk samples, and NPN was determined from a filtrate of whole milk after precipitation with 12% (wt/vol) trichloroacetic acid (17). Protein nitrogen values were converted to protein by multiplying by a factor of 6.38. The fat content was measured by the Gerber method (22). Milk lactose was calculated as the difference between the amount of OM and protein plus fat. The ash content of the feedstuffs, orts, and milk was determined by incineration in an electric muffle furnace at 550°C, and the energy content was determined by adiabatic bomb calorimetry.

The AA composition of the original protein sources and of the residues in the bags after ruminal incubation was determined by HPLC using the Waters Pico-Tag method, which involves precolumn derivatization with phenylisothiocyanate. Protein hydrolysis was performed in 6 *M* HCl in sealed, evacuated tubes at 110°C for 24 h. Cysteine and methionine were determined as cysteic acid and methionine sulfone, respectively, which were obtained by oxidation with performic acid before 6 *M* HCl hydrolysis (7). Tryptophan was not determined.

Milk protein fractions were analyzed by means of SDS-PAGE (PhastSystem™ Pharmacia, Uppsala, Sweden). The SDS-PAGE was performed on 20% homogeneous precast PhastGels™ (Pharmacia) in accordance with the instructions of the manufacturer (file no. 111) (24). The gels were stained automatically in the development unit of PhastSystem™ following fast Coomassie blue staining (file no. 220) (23). Band densities on the SDS gels were quantitated. The gels were scanned using a Bioimate analyzer (3CX′ Bioimage and visage; Millipore Corp., Bedford, MA) according to the Whole Band Analysis Program (18). Standards used were molecular markers (Pharmacia), phosphorylase B (94 kDa), albumin (64 kDa), ovoalbumin (43 kDa), carbonicanhydrase (30 kDa), trypsin inhibitor (20.1 kDa), and lactalbumin (14.4 kDa). Densitometric peak areas from different caseins and from different whey protein fractions were converted to a percentage of the total casein peak area or the total whey protein peak area.

Statistical Analysis

The degradation kinetic parameters of each protein source were estimated using the model of Ørskov and McDonald (21). The effective degradability was calculated as a + (b × c/c + k) where a = quickly degradable fraction, b = slowly degradable fraction, c = rate of degradation, and k = rate of passage of the ingesta. A value of 2.4%/h was used for the latter parameter (13).

The model accounted for variation caused by the protein source in the diet. Results were subjected to an ANOVA in accordance with the general linear models procedure of SAS (29). In goats, DMI is the factor that most determines milk production and composition, regardless of diet type (12, 28). Once it was determined that DMI was not affected by diet, DMI was considered to be independent of the diet (32), and the effect of DMI on milk production and composition (DM, protein, fat, lactose, and energy concentration in milk) was tested as a covariate in the model. When the effect of a covariate factor was not significant ($P > 0.05$), the least squares means were calculated from the model after this term was omitted (32).

To determine the patterns of relationships between specific variables, multivariate factorial analyses were performed using the factor procedure of SAS (29); the algorithm used was PROC FACTOR. The correlation matrix was selected. The methods for the factors extraction and rotation were principal component analysis and varimax, respectively (29). The number of factors derived in each case, in addition to dependence on the essential objective of the analysis, depended on the fraction of the total variance that each of the factors explained as well as on the extent to which each variable defined them. Two multivariate analyses were performed in this way. In the first analyses, the goal was to establish the overall change in the AA composition of protein sources after ruminal incubation. The experimental units were the protein sources both before and after ruminal fermentation. The variables considered were the concentrations of each of the essential amino acids (**EAA**) as well as the total nonessential AA. The second multivariate analysis was performed to establish the pattern of relationships between those variables that defined the nature of the protein sources used and those that defined the amount and composition of the milk protein produced; each goat was considered to be an experimental unit.

RESULTS

Degradability of the Protein Sources

The degradation characteristics of the CP of the protein sources in the rumen are shown in Table 3. The FB had more ($P < 0.05$) of the quickly degradable fraction of CP than did the other three protein sources. In general, the concentrations of slowly degradable fractions of CP were greater ($P < 0.05$) than for the quickly degradable fractions. The maximum values for the fractional rates of degradation were recorded; SFM had a higher ($P < 0.05$) rate than did the other protein sources. The lowest values for effective degradability were observed for CS.

AA Composition of Protein Sources and of RUP Fractions of Protein Sources

Table 4 shows the AA composition of the protein sources and residues in the bags after 72 h of incubation. The different protein sources had similar EAA contents (ranging from 51.0% for FB to 42.2% for CGF). The EAA content of the protein sources was similar before and after ruminal incubation; the maximum change was observed for FB (51.0 vs. 37.6%). To compare the overall changes in the AA composition of each of the protein sources that were caused by the action of the rumen, the first of the multivariate analyses that were described previously was carried out. The results of this multivariate analysis are shown in Table 5 and Figure 1. Two different factors were derived, the first and second accounting for 35.8 and 26.0% of the total variance, respectively. In accordance with the factor loadings, the first factor was mainly determined by the concentration of Arg, Lys, and Phe with positive loading values and by the concentration of nonessential AA and Thr with negative loading values. The second factor was mainly determined by the concentration of His and Thr with positive loading values and by the concentration of Met and Val with negative loading values. In Figure 1, the positions of the different protein sources before and after ruminal degradation and the situations of the different AA are represented in relation to these factors. Two types of change in the AA composition of the protein sources were observed: one in the negative direction of axis 1 (FB and CS) and the other in the positive direction of axis 2 (CGF and SFM). Organization of the feeds in the order of the intensity of change yielded the following: FB, SFM, CS, and CGF.

TABLE 3. Ruminal degradation parameters[1] of the protein sources.

	a		b		c		ED
	(%)						
	$\bar{X}$	SD	$\bar{X}$	SD	$\bar{X}$	SD	
Fababeans	53.50	3.74	54.92	11.35	3.00	1.31	84.0
Sunflower meal	19.30	3.94	79.85	4.47	15.08	2.00	88.2
Corn gluten feed	42.42	5.97	56.32	7.54	5.86	2.50	82.4
Cottonseed	32.13	4.64	43.65	8.79	6.50	3.21	64.0

[1]a = Quickly degradable fraction, b = slowly degradable fraction, c = rate of degradation, and ED = effective degradability.

TABLE 4. Amino acid composition of the protein sources[1] and residues in the bag after 72 h of ruminal incubation.

	FB		SFM		CGF		CS	
	Feed	Residue	Feed	Residue	Feed	Residue	Feed	Residue
	(g/100 g of AA)							
EAA[2]								
Lys	7.5	4.1	3.9	3.2	4.3	5.4	4.7	4.8
His	2.7	3.7	2.2	3.7	2.2	2.1	2.6	3.8
Thr	3.1	7.6	2.6	5.7	3.4	4.5	2.5	4.2
Arg	11.2	5.1	9.3	5.0	6.3	4.5	13.4	6.9
Val	6.8	5.3	6.6	3.7	7.3	8.3	5.9	6.8
Met	0.7	0.9	1.9	1.0	1.6	1.1	1.2	1.5
Ile	4.3	2.5	4.6	4.2	2.6	3.6	3.1	3.7
Leu	9.6	5.3	7.7	10.0	10.4	7.7	6.7	7.4
Phe	5.1	3.1	5.3	3.3	4.1	4.1	6.7	2.9
NEAA[3]								
Tyr	1.3	4.7	0.7	4.5	1.4	0.8	0.9	4.4
Cys	1.0	0.9	1.1	1.6	1.9	1.2	1.4	0.5
Asp	7.6	9.0	9.1	7.1	7.1	10.0	9.3	8.6
Glu	18.1	13.5	24.1	16.5	18.7	13.0	23.6	15.5
Ser	5.6	9.8	4.6	6.1	5.5	7.2	4.8	9.1
Gly	5.3	9.5	7.0	7.2	6.6	11.9	4.8	6.9
Ala	4.7	7.4	4.3	6.7	7.6	5.9	4.3	5.0
Pro	5.4	7.6	5.0	10.5	9.0	7.7	4.1	8.0
Total EAA	51.0	37.6	44.1	39.8	42.2	41.3	46.8	42.0
Total NEAA	49.0	62.4	55.9	60.2	57.8	58.7	53.2	58.0

[1]FB = Fababeans, SFM = sunflower meal, CGF = corn gluten feed, and CS = cottonseed.

[2]Essential AA.

[3]Nonessential AA.

Live Weights of Goats, DMI, and Milk Production and Composition

The mean live weight of the goats was 43 ± 3.1 kg; no change greater than ±1 kg was observed throughout the experimental period. Table 6 shows the mean DMI (grams per day, and grams per kilogram of $BW^{0.75}$ per day), milk production (grams per day), and milk composition (percentage and megajoules per kilogram) in terms of DM, protein, lactose, fat, and energy contents and the mean values for the different protein fractions in the milk produced (percentages). The type of diet did not affect DMI ($P > 0.05$). Only the amounts of lactose were independent of DMI ($P > 0.05$). Type of diet induced changes in milk DM and protein content ($P \leq 0.05$). Thus, milk DM content was higher ($P \leq 0.05$) for goats fed diets containing CGF and CS than for goats fed the diet containing FB. Milk protein content was greater ($P \leq 0.05$) for goats fed the diets containing CGF and FB than for goats fed the diet containing SFM. The α_s-CN percentage was higher ($P \leq 0.05$) for goats fed diets containing FB and SFM than for goats fed diets containing CGF and CS. The β-CN percentage was higher ($P \leq 0.05$) for goats fed the diet containing CGF than for goats fed the diet containing SFM. The κ-CN percentage was greater ($P \leq 0.05$) for goats fed the diet containing FB than for goats fed the diet containing SFM. The serum albumin percentage was higher ($P \leq 0.05$) for goats fed the diets containing FB and SFM than for goats fed the diet containing CS. Finally, the α-LA percentage was higher ($P \leq 0.05$) for goats fed the diet containing SFM than for goats fed the diet containing CGF. The β-LG percentage was higher ($P \leq 0.05$) for goats fed the diet containing CGF than for goats fed the diet containing SFM.

Relationships Between the Amount and Composition of the Milk Protein Produced and the Nature of the Protein Sources Used

For each goat, the protein converted into quickly degradable protein in the rumen (grams per kilogram of $BW^{0.75}$ per day), slowly degradable protein in the rumen (grams per kilogram of $BW^{0.75}$ per day), and effectively degradable protein in the rumen (grams per kilogram of $BW^{0.75}$ per day), and RUP (milligrams per kilogram of $BW^{0.75}$ per day) that was provided by each protein source ingested was calculated as a function of the degradation characteristics estimated for each source. In the same way, the

TABLE 5. Factor pattern for amino acids from protein sources and residues in the bag after 72 h of ruminal incubation.[1]

Variable	Factor matrix	
	Factor 1[2]	Factor 2[3]
Arg	0.8257	–0.2114
Lys	0.7906	0.0763
Phe	0.7134	–0.4438
Ile	0.3880	–0.0848
NEAA[4]	–0.9636	0.2171
His	–0.3204	0.8030
Thr	–0.6223	0.7340
Leu	0.1819	–0.1937
Val	0.1763	–0.6332
Met	–0.3591	–0.8219

Final statistics

Factor	Eigen value	Variance explained	Cumulative variance explained
		(%)	
1	3.5779	35.8	35.8
2	2.5993	26.0	61.8

[1]Results were derived from multivariate analysis.

[2]Expresses mainly the Arg, Lys, and Phe content in the protein sources and residues in the bag.

[3]Expresses mainly the His and Thr content in the protein sources and residues in the bag.

[4]Nonessential AA.

amounts of each EAA (milligrams per kilogram of $BW^{0.75}$ per day) that were provided by the corresponding RUP fraction were calculated from the amino acid composition of residues in the bag. All of these values are shown in Table 7 together with the total milk protein produced (grams per kilogram of $BW^{0.75}$ per day) and the corresponding amounts of the milk protein fractions (grams per kilogram of $BW^{0.75}$ per day).

With the values of all of these variables, those defining the nature of the protein sources used and those defining the amount and composition of the milk protein produced, and with the goal of exploring the relationships among them, a multivariate factorial analysis was carried out, and the results are shown in Table 8. Four different factors were derived, the first, second, third, and fourth accounting for 50.8, 18.9, 11.5, and 10.3% of the total variance, respectively. The first of the derived factors, in accordance with the corresponding factor loadings, was specially defined by the RUP and by its EAA content. These variables appeared with only low factor loadings in the other factors. The amount of milk protein produced and its different casein fractions as well as the quickly degradable protein in the rumen were the variables that most clearly defined the second factor. Factor 3 was primarily determined by the slowly degradable protein in the rumen and by the effectively degradable protein in the rumen. No other variables determined this factor to have a high loading value. Finally, factor 4 was defined by the different milk whey fractions. Figure 2 shows the position of the different variables as well as the area of dispersion for the different protein sources with respect to factors 1 and 2. With respect to factor 1, the different protein sources appeared in different positions; the different protein sources occupied more similar positions with respect to factor 2.

DISCUSSION

Characteristics of the Ruminal Degradation of the Protein Sources

Of the protein sources used, FB were chosen because goats show a distinct preference for this feed. The other three sources were chosen because of their supposed lower degradability. Boza and Ferrando (4) reported that SFM and CGF are good protein concentrates; CS is a good source of bypass protein as well as an excellent energy source. Results obtained from

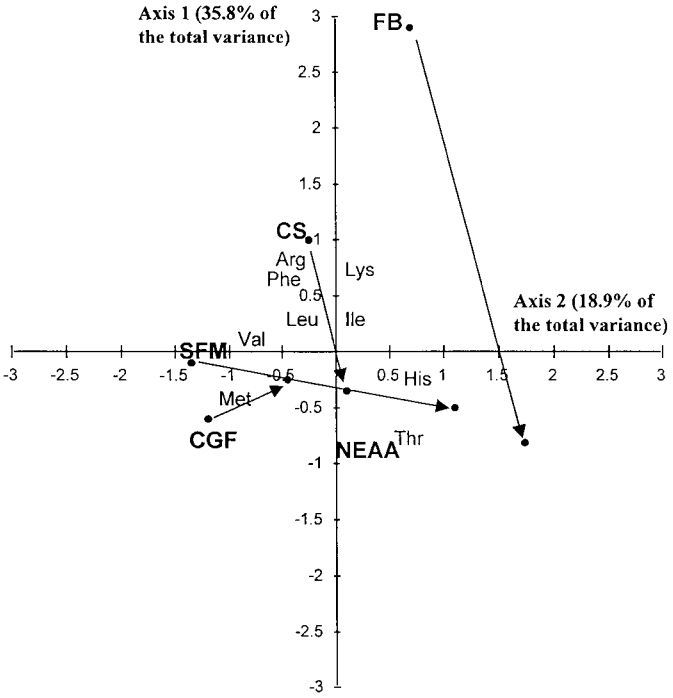

Figure 1. Effects of ruminal degradation on the factor pattern of the AA profile of the protein sources. CS = Cottonseed, FB = fababeans, CGF = corn gluten feed, SFM = sunflower meal, and NEAA = nonessential AA. Point at end of arrow represents the factor pattern of the AA profile of residue remaining in the bag.

TABLE 6. Milk production and composition by goats fed diets differing in protein source.

	Diet[1]					*P*	
	FB	SFM	CGF	CS	RSD[2]	Covariate	Diet
DMI, g/d	1157	1064	1064	1155	254.74	...	NS[3]
DMI, g/kg of $BW^{0.75}$ per d	69.3	61.5	65.2	68.4	15.17	...	NS
Milk production, g/d	1071	1153	971	1098	269.70	***	NS
Milk composition							
DM, %	15.21[b]	15.52[ab]	16.40[a]	16.33[a]	0.71	***	*
Protein, %	3.25[a]	2.87[b]	3.50[a]	3.18[ab]	0.31	**	**
Fat, %	6.00	6.57	6.32	6.61	0.10	***	NS
Lactose, %	5.12	5.71	5.77	5.63	0.67	NS	NS
Energy, MJ/kg	3.9	4.1	4.0	4.1	0.29	**	NS
Protein fractions							
Casein, % of protein	70.5[b]	70.7[b]	80.9[a]	74.2[b]	7.16	...	*
Whey protein, % of protein	29.5[a]	29.3[a]	19.1[b]	25.8[a]	7.11	...	*
SA,[4] % of Whey protein	14.6[a]	14.3[a]	11.9[ab]	11.0[b]	2.30	...	*
α-LA, % of Whey protein	27.1[ab]	29.8[a]	23.9[b]	26.8[ab]	3.70	...	*
β-LG, % of Whey protein	58.3[ab]	56.0[b]	64.2[a]	62.2[ab]	5.97	...	*
α_3-CN, % of Casein	35.4[a]	37.4[a]	26.1[b]	28.8[b]	3.93	...	**
β-CN, % of Casein	56.0[b]	56.7[ab]	66.1[a]	63.6[ab]	6.42	...	*
κ-CN, % of Casein	8.7[a]	5.9[b]	7.8[ab]	7.6[ab]	1.66	...	*

[a,b]Means within a row with unlike superscripts differ ($P \leq 0.05$).

[1]Twenty percent of protein was provided by fababeans (FB), sunflower meal (SFM), corn gluten feed (CGF), and cottonseed (CS).

[2]Residual standard deviation.

[3]$P > 0.05$.

[4]Serum albumin.

*$P \leq 0.05$.

**$P \leq 0.01$.

***$P \leq 0.001$.

this study for the characteristics of ruminal degradation showed that FB, SFM, and CGF had similar effective degradation. Romero et al. (25) worked with goats fed diets containing soybean meal or CGF as the protein source; those researchers noted a higher milk production by goats fed the CGF than that by goats fed soybean meal. Those researchers (25) attributed this result to the higher content of RUP that reached the duodenum. Corn protein has a lower degradability than does soybean meal protein.

AA Composition of the Protein Sources Used and of Their RUP Fractions

In spite of the Rulquin and Verité (26) opinion that feed AA profiles can be taken as a first guide to estimate the AA profiles of RUP, other researchers (8, 9, 15) reported data from which it can be deduced that the use of the original AA profile of one protein source to predict EAA available for absorption is not accurate because the change that is undergone differs according to the protein source, such as was inferred from this study. The AA profiles of the residues of feeds subjected to ruminal fermentation shows that Arg is one of the amino acids that is most sensitive to such fermentation. As a result, its concentration decreased (8). In contrast, the amounts of Ile and Phe might have increased, and the amount of Lys might have decreased (9, 15). In the present study, a marked decrease was noted in the concentration of Arg in the four protein sources. At the same time, Phe and Lys decreased in the protein sources, except in CGF and CGF and CS, respectively; Thr increased in all four of the protein sources. After examination of the effect of ruminal incubation on the AA profile by multivariate analysis, Rulquin and Verité (26) concluded that these differences were small in comparison with the differences that continue to exist among feeds. In the present study, when a multivariate analysis was carried out that was similar to that by Rulquin and Verité (26), the results showed clearly that the overall change of the AA profile depended on the feeds (Figure 1). The AA profiles of FB and SFM were very different before ruminal fermentation. However, the AA compositions of their corresponding RUP fractions were much closer. Something similar occurred with the CS and CGF. The degree of modification brought about by ruminal fermentation of the AA profile of each protein source is given by the magnitude of the vector that joins the position of each one before and after ruminal fermentation.

Milk Production and Composition

The milk production by goats fed diets with practically equal energy and N contents is known to be dependent on the intake of the diet. Hadjipanayiotou and Morand-Fehr (12) reported that feed intake is the chief factor that determines milk production by goats. The correlation between the energy intake and milk production halfway through lactation is 0.83. In the present study, analyses of milk production, considering rates of DMI as a covariance factor, yielded results that indicated that production depended on intake. No additional differences were detected as a result of the type of diet.

In goats, energy intake is the factor that is the most significant determinate of milk composition (28). With this in mind and with the goal of determining the effect of the different types of diet on milk composition, the values of different parameters that are indicative of composition were analyzed statistically; the DMI in each case was used as the covariance factor. In the lactating ruminant, less degradable proteins are used to increase the milk protein content as long as the animal has the potential to increase milk protein (5). However, studies carried out on goats to investigate this effect (11, 19) have given different and, at times, contradictory results. Morand-Fehr et al. (19) have reported that in the majority of cases in which isoenergetic diets with the same N content are used, the protein and casein contents of the goat milk do not appear to be particularly sensitive to changes in the protein source of the diet. The same result was found to be true for milk fat content. However, the effect of a change in the protein source of the diet on the protein content of caprine milk has always been analyzed by comparing alternative protein sources with soybean protein. Results could vary with the nature of the protein that the less degradable protein replaces. Here the best results were obtained when goats were fed the diet contain-

TABLE 7. Diet and overall means of the variables that define the nature of the protein sources and the amount and composition of the milk protein produced.

	Diet[1]					
	FB	SFM	CGF	CS	$\bar{X}$	SEM
Daily intake per kg of $BW^{0.75}$						
QRDP,[2] g	1.40	0.49	0.92	0.72	0.89	0.10
SRDP,[3] g	1.43	2.04	1.21	0.98	1.40	0.12
ERDP,[4] g	2.19	2.26	1.77	1.52	1.93	0.14
RUP, mg	23.5	69.1	97.0	347.8	134.8	35.10
EAA[5] in RUP, mg						
Lys	0.96	2.21	5.23	16.70	6.28	1.75
His	0.87	2.56	2.33	13.22	4.80	1.38
Thr	1.79	3.94	4.36	14.61	6.18	1.44
Arg	1.20	3.46	4.36	24.00	8.26	2.55
Val	1.25	2.56	8.04	23.65	8.88	2.49
Met	0.21	0.69	1.07	5.22	1.80	0.56
Ile	0.59	2.90	3.49	12.87	4.96	1.33
Leu	1.25	6.91	7.47	25.74	10.35	2.63
Phe	0.73	2.28	3.97	10.09	4.27	1.02
MP[6] and MP Fractions, g/kg of $BW^{0.75}$						
MP	1.96	1.51	1.92	1.90	1.82	0.14
α_s-CN	0.47	0.40	0.41	0.44	0.43	0.03
β-CN	0.79	0.59	1.01	0.90	0.86	0.09
κ-CN	0.14	0.07	0.13	0.10	0.11	0.01
SA[7]	0.07	0.06	0.05	0.05	0.06	0.01
α-LA	0.16	0.14	0.08	0.13	0.13	0.01
β-LG	0.32	0.26	0.25	0.29	0.28	0.02

[1]Twenty percent of protein was provided by fababeans (FB), sunflower meal (SFM), corn gluten feed (CGF), and cottonseed (CS).

[2]Quickly degradable protein in the rumen.

[3]Slowly degradable protein in the rumen.

[4]Effectively degradable protein in the rumen.

[5]Essential AA.

[6]Milk protein.

[7]Serum albumin.

ing CGF. In addition to the higher milk protein production by goats fed this diet, although not significantly higher than that produced by goats fed the diet containing FB, the milk casein percentage was higher than that produced by goats fed the other diets. This increase was closely linked to the β-CN percentage. The milk production by goats fed the diets containing SFM had the lowest protein content. As is discussed later, these results were due to the effect of the nature of the protein source used on the amount and composition of the milk protein produced.

Nature of the Protein Sources and Amount and Composition of the Milk Protein

The new systems used to evaluate the protein in the feeds of lactating ruminants (1, 20) suggest that animals need to absorb a certain amount of protein to ensure that there is a certain supply of RDP and RUP. However, with greater frequency, opinions and experimental results (5, 15, 26, 27, 31) indicate that it is necessary to take into account first, the kinetics of the ruminal degradation of the protein supplied and second, the AA profile of the RUP fractions of the protein supplied. These results suggest the necessity of taking into account the process of milk protein production from the protein consumed by the animal, which is a multivariate process.

When the effects that the kinetics of ruminal degradation of a protein can have on the use of the protein supplied for milk production are considered, it is necessary to examine the values estimated here for the different parameters considered in the model of ruminal degradation developed by Ørskov and McDonald (21) as a function of the protein source

TABLE 8. Variables that define the nature of the protein sources and the amount and composition of the milk protein produced. Results were derived from multivariate analysis.

	Factor matrix			
Variable	Factor 1[1]	Factor 2[2]	Factor 3[3]	Factor 4[4]
Ile	0.9977	0.0522	0.0165	0.0319
Leu	0.9972	0.0411	0.0386	0.0255
RUP	0.9963	0.0764	0.0048	0.0327
Thr	0.9946	0.0742	0.0505	0.0439
Met	0.9940	0.0651	–0.0273	0.0588
Arg	0.9926	0.0542	–0.0178	0.0745
Lys	0.9912	0.1163	–0.0341	0.0063
Phe	0.9897	0.0266	–0.0061	0.0983
His	0.9895	0.1136	0.0102	–0.0276
Val	0.9864	0.1353	–0.0493	–0.0109
κ-CN	–0.0922	0.9488	–0.0643	0.1659
β-CN	0.2993	0.8938	0.0795	–0.0486
MP	0.2704	0.8735	0.2240	0.2784
QRDP	–0.0829	0.7322	0.3372	0.0494
α_s-CN	0.3247	0.6414	0.4961	0.3727
SRDP	–0.0945	0.0821	0.9514	0.0289
ERDP	0.0116	0.3899	0.9087	0.0315
α-LA	0.0341	0.1413	0.3072	0.8560
β-LG	0.0533	0.3413	0.0927	0.8123
SA	0.0160	–0.0198	–0.1787	0.6337

Final statistics

Factor	Eigen value	Variance explained	Cumulative variance explained
		(%)	
1	10.1565	50.8	50.8
2	3.7736	18.9	69.7
3	2.2953	11.5	81.2
4	2.0690	10.3	91.5

[1]Expresses the RUP and its essential AA content, weakly associated with milk protein (MP) and with its α_s-CN and β-CN fractions.

[2]Expresses the quickly degradable protein in the rumen (QRDP), mainly associated with MP and with its different casein fractions.

[3]Expresses the slowly degradable protein in the rumen (SRDP) and effectively degradable protein in the rumen (ERDP), weakly associated with α_s-CN and α-LA.

[4]Expresses the different whey protein fractions, α-LA, β-LG, and serum albumin (SA).

used (Table 3). This examination reveals that it is possible to derive similar values for effective degradability from quite different parameters. In fact, the effective degradabilities estimated for the protein from FB, SFM, and CGF were very similar despite the fact that the values for the quickly degradable fraction in the rumen, the slowly degradable fraction in the rumen, and the rate of degradation were quite different. If the AA composition of the RUP fractions of the protein sources (Table 4) is considered, it can be seen that the proportions of some AA were similar (His, Met, and Phe) and those for others were different (Thr, Val, and Leu), resulting in nearly identical total proportions of EAA at the same time for the different protein sources. Because of these results, it is worthwhile to determine which aspects of the composition or nature of the protein sources used most affect the amount and even the composition of the milk protein that is produced.

With regard to a determined physiological process, when the values of a series of relevant variables are available, multivariate statistical analysis is an appropriate methology to identify biological similarities among them. Various researchers (6, 30) have indicated how the results obtained in this way acquire statistical as well as biological significance. In our case, a multivariate factorial analysis was carried out with the values of all derived variables: those defining the nature of the protein sources used and those defining the amount and composition of the milk protein produced. Because, in this type of analysis, the factors represent new variables that are not correlated with each other (6), the identification of groups of variables that behaved differently and independently was possible. With regard to the variables that define the nature of the protein sources, the results highlighted the different behaviors of first, the RUP, second, the quickly degradable protein in the rumen, and third, the slowly degradable protein in the rumen and the effective protein degradable in the rumen. These variables intervened only in the definition of factors 1, 2, or 3, respectively. The quickly degradable protein in the rumen was the fraction most closely related to the milk protein produced, in particular to its casein fractions. To a much lesser extent, the RUP fraction, regardless of its EAA composition, also was related to milk protein with regard to α_s-CN and β-CN fractions. No protein supplied fraction was related to any whey protein fraction. So the different whey protein fractions were the most independent of the nature of the protein source. As Chatfield and Collins (6) reported, two different goals could be realized with this type of multivariate analysis: first, examination of the pattern of relationships that existed among the considered variables and, second, the determination of whether, according to the results obtained, it was possible to distinguish among the protein sources. Figure 2 separates and classifies the feeds, especially with respect to the first factor. In this way, it may be possible to establish a hierarchy among the four protein sources: CS, CGF, SFM, and FB.

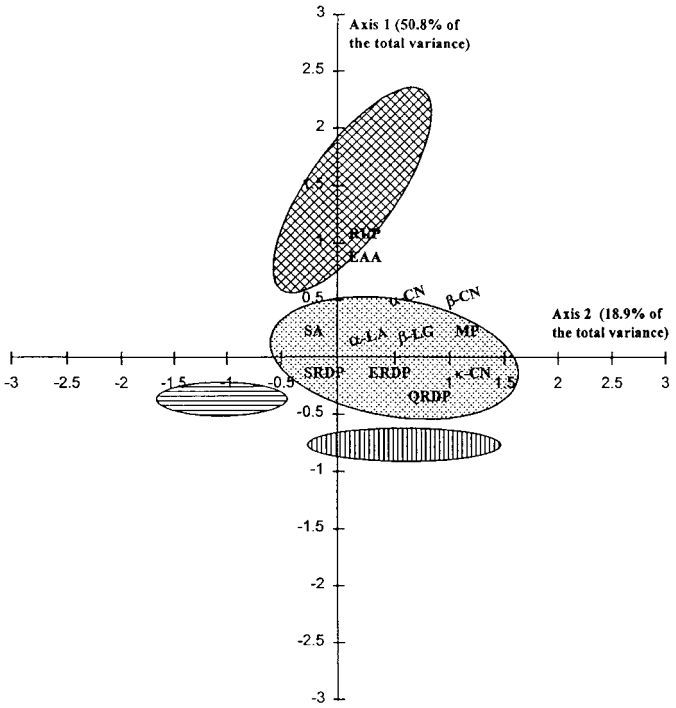

Figure 2. Variables that define the nature of the protein sources and the amount and composition of milk protein (MP) as related to source of protein used. First and second factors in the multivariate analyses were used to plot the variables. QRDP = Quickly degradable protein in the rumen, SRDP = slowly degradable protein in the rumen, ERDP = effectively degradable protein in the rumen, EAA = essential AA, and SA = serum albumin. Dispersion area of protein sources: fababeans (vertical lines), sunflower meal (horizontal lines), corn gluten feed (dotted pattern), and cottonseed (cross-hatched pattern).

With regard to the significance of the quickly degradable protein in the rumen as deduced here, it is worth bearing in mind the observations of Chandler (5), who reported that to satisfy the protein requirements of lactating ruminants best, it is necessary to identify those protein sources that will be quickly degraded first (i.e., the N that is supplied to the animal in the form of microbial protein). Chandler (5) also pointed out that the supply of this microbial protein could be balanced using protein sources that are resistant to ruminal degradation. The animal would thus acquire an additional supply of N. The significance of this additional N for the lactating ruminant appears to depend on both the amount of N supplied and the AA profile (5, 15, 26, 27, 31). In our

study, the possible supplementary N achieved from the RUP fraction was established by the amount supplied, regardless of its EAA composition. In this sense, one should consider the comments of Schwab et al. (31) who observed that the EAA with the most limiting effect on milk production are Lys and Met. In the present study, the concentrations of these two AA were similar in the RUP fractions from each source. Furthermore, as can be observed from Figure 1, all of the undegradable fractions were more similar in amino acid composition than were protein sources prior to ruminant degradation.

CONCLUSIONS

The composition of the milk produced by Granadina goats appears to be sensitive to diets with 20% of the protein provided by FB, SFM, CGF, and CS. With regard to the milk protein produced, the protein sources used consisted of different and independent entities. The quickly degradable protein in the rumen was most closely related to the milk protein produced. To a much lesser extent and regardless of the EAA composition, the RUP fraction was also related to milk protein production.

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