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Milk qualitative and quantitative characteristics, metabolic profile and rumen pH and NH_3 concentration in grazing goats fed with two types of concentrate

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SUMMARY – The aim of this study was to investigate the effect of two types of concentrates characterized by different degradation (starch and nitrogen rapidly degradable [PBC] and less rapidly degradable [PMB]) compared with the group not supplemented (P) on milk quantitative and qualitative (protein, fat, lactose, urea, saturated [SFA], monounsaturated [MUFA] and polyunsaturated [PUFA] fatty acids [FAs], CLA, C18:1t, ω-3, ω-6, thrombogenic index, atherogenic index) characteristics in lactating goats grazing on natural pasture. Goats were also monitored for rumen pH and ammonia (NH_3) concentration and for their metabolic profile (NEFA, glucose, cholesterol, protein, urea, albumin and globulin). Concentrate supplementation increased milk yield in groups PBC, while in group P and PMB milk yield decreased, respectively 27% and 9%. Milk protein, fat, lactose and urea were not affected by the feeding regimen. CLA and C18:1 trans values increased by 47% and 39% in P and by 45% and 43% in PMB compared to PBC group, respectively. No differences in SFA, MUFA, PUFA fatty acids, ω-3, ω-6 FAs and thrombogenic index were found between PBC and PMB groups. The dietary treatments had a significant effect on the metabolic profile of the goats. Non-sterified fatty acids (NEFA) and cholesterol concentrations showed the highest level in group PBC. Group PMB presented the lowest NEFA and glucose concentration. Plasma urea levels were significantly higher in group P indicating an inefficient use of ruminal nitrogen. Total protein and globulin concentrations were higher in group PMB, while the maximum level of albumin was observed in group P. The maximum decrease in pH was found after two hours in PBC and after four hours in PMB. The ruminal NH_3 concentration was the greatest for PBC at any time of ruminal fluid sampling. Results indicate that, under the grazing conditions, PMB dietary treatment improved milk quality, while milk yield was not significantly lower than PBC; also, in PMB group plasma metabolites showed better conditions of animals.

Keywords: Milk quality, milk quantity, metabolic profile, degradation of concentrate, grazing goat.

RESUME – *"Effet de deux types de complémentation sur la production et la qualité du lait, le profil métabolique, le pH et la concentation de NH_3 dans le rumen de chèvres au pâturage". L'objectif de cette expérience a été d'étudier l'effet de différents types de complémentation (concentrés à vitesse de dégradation plus élevée [groupe PBC] ou moins élevée [groupe PMB] de l'amidon et de l'azote par rapport à un groupe sans intégration [groupe P]) sur la production et la qualité du lait (matières azotées totales [MAT], taux butyrique, lactose, urée, acides gras saturés [SFA], acides gras mono-insaturés [MUFA], acides gras polyinsaturés [PUFA], CLA, C18:1t, ω-3, ω-6, indice thrombogène et indice athérogène) chez des chèvres au pâturage. Sur deux chèvres par groupe, ont été déterminées les valeurs du pH et la concentration de NH_3 au cours des 24 heures. En revanche, on a déterminé sur chaque groupe les paramètres du profil métabolique (acides gras non estérifiés [AGNE], glucose, cholestérol, protéine, urée, albumine et globuline). La complémentation distribuée a augmenté de manière significative la production de lait dans le groupe PBC, tandis que, au contraire, elle l'a diminuée, respectivement de 27% et de 9%, dans les groupes P et PMB. La teneur en MAT, le taux butyrique, le lactose et l'urée n'ont pas été influencés par le régime alimentaire. La teneur en CLA et C18:1 trans est augmentée, respectivement, de 47% et de 39% dans le groupe P et de 45% et de 43% dans le groupe PMB par rapport au groupe PBC. Aucune différence n'a été observée pour la teneur en acides gras SFA, MUFA, PUFA, ω-3, ω-6 et indice thrombogène entre les groupes PBC et PMB. Le régime alimentaire, par contre, a montré un effet considérable sur le profil métabolique des chèvres. La concentration la plus élevée en AGNE et cholestérol a été observée dans le groupe PBC. Dans le groupe PMB ont été enregistrées les valeurs les plus faibles en AGNE et glucose. Les valeurs élevées d'urée plasmatique relevées dans le groupe P indiquent une utilisation insuffisante de l'azote au niveau du rumen. Les valeurs les plus élevées de protéine totale et de globuline ont été observées dans le groupe PMB, tandis que la valeur la plus élevée d'albumine a été observée dans le groupe P. La baisse maximale du pH a été observée après deux heures dans le groupe PBC et après quatre heures dans le groupe PMB. La concentration de NH_3 dans le jus de rumen du groupe PBC a résulté, pour tous les temps considérés, plus élevée par rapport aux autres groupes. Les résultats obtenus indiquent que, pour les chèvres au pâturage, le régime PMB améliore la qualité du lait, tandis que la production de lait n'est pas significativement inférieure avec le régime PBC ; en outre dans le groupe PMB les métabolites plasmatiques indiquent de meilleures conditions des animaux.*

Mots-clés : *Production et qualité du lait, profil métabolique, dégradabilité du concentré, chèvres au pâturage.*

Introduction

The objective to optimise the quantitative and qualitative milk production, in order to meet the health and nutritional characteristics (CLA, ω-3 fatty acids, thrombogenic and atherogenic indexes) demanded by the consumer, implies the use of nutrient supplementation especially in grazing systems that rely on pasture utilization. Traditionally, goats breed in southern Italy are not supplemented with concentrates, although in recent years this practice has become more common (Rubino *et al.,* 1995). Herbage quality and availability, the type and level of concentrate may contribute to change milk fatty acid composition of goat milk as well as milk yield. The effect of different types of concentrates, characterized by different rumen starch and nitrogen degradability, has been studied in goats reared indoors and fed a total mixed ratio (TMR) (Schmidely *et al.*, 1999). However, to the best of our knowledge, studies on the effects of feed supplementation in grazing dairy goats reared under grazing conditions are lacking. Of particular interest would be the study of the different combinations of pasture and concentrates, as it would affect rumen environment and the metabolic profile of the animals. The aim of this study, thus, was to investigate the effect of feeding grazing dairy goats with two types of concentrates characterized by rapid or less rapid rumen degradability of starch and nitrogen on milk yield and its composition. In particular, milk was assessed for protein, fat, lactose and urea content, saturated fatty acids (SFA), monounsaturated fatty acids (MUFA), polyunsaturated fatty acids (PUFA), conjugated linoleic acids (CLA), C18:1t, ω-3, ω-6, thrombogenic index and atherogenic index. Goats were also monitored for rumen pH and NH_3 concentration and for their metabolic profile (non-esterified fatty acids [NEFA], glucose, cholesterol, protein, urea, albumin and globulin).

Material and methods

In spring (April), 45 Red Syrian goats (92 ± 7 DIM) were randomly allocated to three different groups and fed with three different diets: pasture plus 550 g DM/d of barley and chickpeas (rapidly degradable starch and nitrogen, PBC; 16.2% CP, 3.1% EE, 15.8% NDF), pasture plus 550 g DM/d of maize and broad-beans (less rapidly degradable starch and nitrogen, PMB; 15.4% CP, 3.3% EE, 15.0% NDF) and pasture without concentrate supplementation (P; 17.8% CP, 3.1% EE, 40.0% NDF). The different rumen degradability of PMB and PBC diets has been previously reported (Claps *et al.,* 2000). The goats received the nutritional treatment for 4 weeks and milk, blood and rumen sampling started 10 days after the beginning of the administration of the different diets. Milk yield was recorded and milk and blood samples were collected from all animals every two weeks. Milk samples were analysed for fat, protein and lactose content by Milko-Scan (Foss-Electric). Milk urea was assayed after protein precipitation with trichloroacetic acid and neutralisation. A duplicate set of milk samples was stored at -20°C until fatty acid analysis. Milk lipids were extracted with chloroform and methanol and fatty acids (FAs) were converted into methyl esters and separated by gas chromatography (Varian 3800 with CP 8410 auto injector) equipped with a FID detector and a 60 m x 0.25 mm (id) cyanopropyl polysiloxane (DB 23, J & W) fused silica capillary column. Operating conditions have been described previously (Di Trana *et al.,* 2004). Each fatty acid was identified with reference to the retention time of the standards (Sigma and Larodan). Results of fatty acids were expressed as percentage of fatty acid methyl ester (FAME). The thrombogenic index and atherogenic index of milk were calculated according to Ulbricht and Southgate (1991). Blood was centrifuged and plasma was immediately assayed for glucose and NEFA (Randox, FA 115). Plasma was then stored at -20°C until assayed for cholesterol, total protein, albumin and urea, which were determined using an autoanalyzer (Polimak PM M10/2) with commercial kits. Globulin concentration was calculated as differences between total protein and albumin. Within each group, two goats were fitted with a ruminal cannulae in order to assess rumen pH and NH_3 values which were measured at 5 min., 2h, 4h, 8h, 12h and 24h from feeding time. Rumen NH_3 concentration was measured by a pH-meter specific electrode, according to official methods of analyses (AOAC, 2000). Statistical treatment of the data was performed using SYSTAT statistical package (Systat, 1992). Milk yield, milk composition, fatty acid composition, blood parameters, ruminal pH and ruminal NH_3 were analysed by ANOVA procedure including the effect of feeding regimen. The comparison between means was performed using Fisher’s LSD test.

Results and discussion

Milk yield was significantly influenced by feeding regimen (P<0.05). The highest milk yield was detected in Group PBC, and its values were 27% and 9% higher than those observed in Groups P and PMB (Table 1). In agreement with our observation, an increase of milk yield has been found in Alpine goats consuming the rapidly degraded dry diet (Schmidely *et al.,* 1999). The nutritional treatments adopted did not significantly modify milk composition. Milk fat tended to be higher in goats fed at pasture (Group P) than in goats fed supplemented (Groups PMB and PBC). Milk urea showed high mean value in Group P, nevertheless no significant difference was detected between groups (P>0.05).

Table 1. Effect of feeding regimen on milk yield and milk composition of lactating goats (means ±SE)

Parameter		Feeding regimen (FR)			SE	Effect of FR
		P	PMB	PBC		
Milk yield	g/d	$1{,}075^{b}$	$1{,}351^{ba}$	$1{,}483^{a}$	140.1	*
Fat	%	3.3	3.0	2.9	0.27	ns
Total protein	%	2.9	3.0	2.8	0.10	ns
Lactose	%	4.7	4.9	4.8	0.12	ns
Urea	mmol/l	9.8	9.1	9.3	0.30	ns

Means within row with different small superscripts differ at P<0.05; *:P<0.05; ns: not significant.

Feeding regimen significantly affected (P<0.001) milk CLA and C18:1trans concentrations (Table 2). CLA and C18:1trans levels were higher in Groups P and PMB compared to Group PBC. The percentage of SFA and MUFA was affected (P<0.001) by feeding regimen, while PUFA didn't show differences between groups. SFA were lower in P than in PBC Group, while MUFA were significantly higher in P than PBC Group. The highest mean level of PUFA was detected in Group P followed by Groups PMB and PBC. Feeding regimen influenced total ω-3 fatty acids (P<0.001) and total ω-6 content (P<0.05).

Table 2. Effect of feeding regimen on milk fatty acid composition (% FAME) and on atherogenic and thrombogenic indexes (mean ± SE)

Parameters	Feeding regimen (FR)			SE	Effect of FR
	P	PMB	PBC		
CLA	0.857^{A}	0.846^{A}	0.581^{B}	0.045	***
C18:1trans	1.800^{A}	1.843^{A}	1.286^{B}	0.163	***
Saturated fatty acids (SFA)	69.672^{A}	70.889^{AB}	71.770^{B}	0.741	***
Monounsaturated fatty acids (MUFA)	25.209^{A}	24.202^{AB}	23.526^{B}	0.617	***
Polyunsaturated fatty acids (PUFA)	5.118	4.909	4.703	0.198	ns
ω-3	1.052^{A}	0.814^{B}	0.824^{B}	0.046	***
ω-6	2.331^{a}	2.378^{ab}	2.575^{b}	0.123	*
Atherogenic index	2.37	2.63	2.59	0.072	ns
Thrombogenic index	2.67^{A}	2.88^{B}	3.03^{B}	0.054	***

Means within row with different small superscripts differ at P<0.05; ***:P<0.001; **:P<0.01; *:P<0.05; ns: not significant.

The highest content of ω-3 was found in milk from goats fed at pasture (Group P), and no differences emerged between groups which utilized supplement rapidly degradable (PBC) and less rapidly degradable (PMB). On the contrary, the lowest content of ω-6 was observed in Group P and its highest in PBC (P<0.05). The atherogenic index was not affected by feeding regimen however its lowest value was found in Group P. The effect of feeding regimen was evident on the thrombogenic index, in fact a significant (P<0.001) decrease was observed in milk of goats fed at pasture (P). No differences were detected in concentrate-supplemented goats. Pasture appears to play a crucial role in determining milk FAs composition. However, in concentrate-supplemented goats, milk FAs profile of group PMB seems to have better characteristics in terms of human health perspective. We previously observed that the pasture was rich in PUFA and especially in ω-3 PUFA such as α-linolenic acid (Di Trana *et al*., 2004). They are the precursor of longer chain n-PUFA such as CLA, EPA and DHA fatty acids (Schmidely and Sauvant, 2001) the latter ones being considered important since they reduce the risk of coronary heart diseases (Sargent and Henderson, 1995). This observation could partially explain differences in milk FAs profiles between groups as we previously detected differences in herbage intake (Claps *et al*., 2001), in grazing behaviour and also in the synchrony of herbage and concentrate degradation in dairy goats fed on a natural pasture or concentrate-supplemented (Claps *et al*., 2000). The effect of the feeding regimen on the metabolic profile of the goats is shown in Table 3. NEFA levels were significantly lower in PMB than in PCB Group (P<0.05). Plasma glucose concentration was significantly (P<0.05) affected by feeding regimen, and its highest values were found in Group P. Total protein (P<0.01) and globulin (P<0.01) concentration were significantly higher in PMB and PBC than in P Group. Plasma urea concentration was significantly higher in P than PBC and PMB Groups (P<0.01). No effect of feeding regimen was noted on plasma levels of cholesterol and albumin. All parameters fall within the range reported for goat species (Kaneko, 1989), and for their physiological state (Ridoux *et al.,* 1981). A similar energetic profile was detected in Red Syrian goats reared in extensive condition (Di Trana and Celi, 1997). The highest level of NEFA observed in Group PBC, associated to its high levels of milk yield may indicate a physiological lipo-mobilisation to sustain milk production. The increase of total protein in Groups PMB and PBC was due to an increase in globulin, however, as already stated before this parameter was in the standard range of the species, suggesting a good metabolic status of the goats. Finally, the lack of differences between groups for albumin and cholesterol levels demonstrated the consistency of high feeding regimen with good liver functionality. The increase in plasma concentration of urea in Group P may indicate inefficient use of ruminal nitrogen (Brun-Bellut *et al.,* 1991) due to energy deficiency during spring season (Masson *et al.,* 1991).

Table 3. Effect of feeding regimen on metabolic profile of lactating goats (mean ± SE)

Parameter		Feeding regimen (FR)			SE	Effect of FR
		P	PMB	PBC		
NEFA	(µmol/l)	226^{ab}	198^{b}	317^{a}	33.1	*
Glucose	(mmol/l)	2.94^{a}	2.40^{b}	2.67^{ab}	0.15	*
Cholesterol	(mmol/l)	1.73	1.70	1.90	0.07	ns
Total protein	g/l	62.70^{A}	76.26^{B}	74.16^{B}	2.88	**
Albumin	g/l	32.69	31.22	31.30	0.71	ns
Globulin	g/l	30.00^{A}	45.04^{B}	42.86^{B}	2.86	**
Urea	(mmol/l)	11.63^{Aa}	9.06^{b}	8.81^{B}	0.64	**

Means within row with different capital or small superscripts differ respectively at P<0.01 and P<0.05; **:P<0.01; *P<0.05; ns: not significant.

Rumen fluid pH was affected by feeding regimen (Fig. 1). Rumen pH did not change in Group P, during the monitoring period (24h), and its values were consistently higher than those observed in Groups PMB and PBC. A similar result was recorded for dry pregnant goats receiving a mixed diet with starch and nitrogen rapidly and less rapidly degradable (Schmidely *et al*., 1996). Natural pastures have a lower content of non-structural carbohydrates and this may have prevented a large decrease in pH unit. After 2h from the administration of the diet, rumen pH decreased in Groups PMB and PBC.

Rumen pH values remained low until 12h and then returned to basal level after 24h. The high availability of non structural carbohydrates, substrates for ruminal microflora, in groups that received supplement (PMB and PBC) might be linked to the decrease of ruminal pH in these groups. Ruminal NH_3 concentration is reported in Fig. 2. The mean concentration of NH_3 in all groups was higher than 50mg/l, this value has been considered minimal for microbial protein synthesis (Schmidely *et al.,* 1996), also this could be due to the elevated capacity for urea recycling in goats (Devendra, 1978). Group P was characterized by small changes in rumen NH_3 concentration. A slight increase in ruminal NH_3 was observed in Group PBC after 2h, but then its values returned to their basal levels and remained fairly constant. In Group PMB we observed an opposite trend, with NH_3 levels that immediately decreased after concentrate administration (5 min) and then slowly returned to pre-feeding levels. The significant difference ($P<0.05$) in rumen NH_3 observed between Groups PBC and PMB after 2h could be due to the higher nitrogen degradability of the concentrates (Claps *et al.*, 2000) which resulted in a higher rumen NH_3 content in Group PBC.

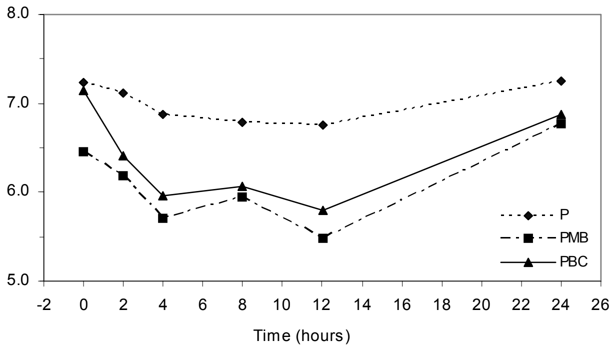

Fig. 1. Trend of rumen liquid pH in different feeding regimes.

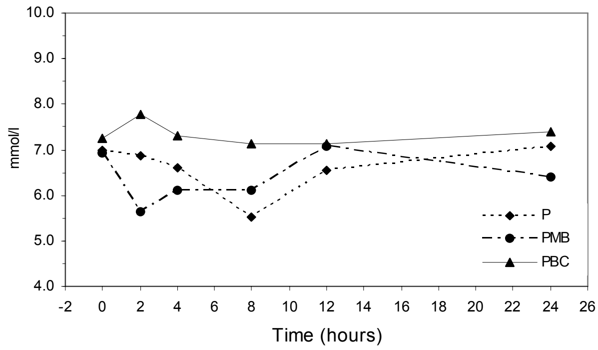

Fig. 2. Trend of rumen liquid NH_3 in different feeding regimes.

Conclusions

The results of this study indicate that pasture herbage play a fundamental role in determining milk FAs composition. The goats fed without supplementation showed a high level of plasma urea that might be linked to the inefficient use of ruminal nitrogen. Finally, during spring season maize and broad beans supplementation are a useful means to optimise milk production and FAs composition in terms of human health perspective, determining also, a good metabolic status and liver functionality in the goats.

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