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Effect of partial replacement of concentrate with feed blocks including tomato wastes from greenhouse horticulture on methane and milk production and milk composition in goats

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Abstract. The aim of the present work was to study, in dairy goats, the effect of replacing 35% of concentrate in the diet with feed blocks including tomato fruits wastes on CH₄ emission, milk yield and fatty acid and amino acid profiles. Eight Granadina goats (39.6 ± 1.89 kg BW) in the middle of the third lactation were used, and a two-periods trial was carried out following a crossover design. In each period, 4 animals randomly received 1.0 kg of alfalfa hay (AH) plus 1 kg of concentrate (diet AC) and the other 4 received 1.0 kg AH plus 0.65 kg of concentrate plus tomato feed blocks *ad libitum* (diet ACB). The amounts of AH and concentrate supplied to the animals fed diet AC were sufficient to allow daily milk production of up to 2 kg per goat (Aguilera et al., 1990). The feed block was supplied *ad libitum* resulting in average intakes of 208 ± 31.8 g of DM/animal/d. Each period included 25 d for adaptation to diet and 8 d for sampling the 3 last for CH₄ measurement in chambers. The ACB diet resulted in a 38.3% reduction (P = 0.001) in CH₄ emissions, and increased (P ≤ 0.048) average proportions of linolenic, linoleic and total polyunsaturated fatty acids in milk. The amino acid profile and concentration were unaffected by diet, but the proportion of phenylalanine was lower (P = 0.039) in ACB milk compared to the control diet. It is concluded that feed blocks based on tomato fruits wastes could replace 35% of the concentrate in dairy goats diet without detrimental effects on milk production and fatty acid or amino acid composition. Overall, diet including tomato feed blocks promoted the production of milk with healthier fatty acid profile and reduced feeding cost and the environmental damage caused by CH₄ emissions, compared with a conventional diet.

Keywords. Goat – Feed blocks – Tomato – Methane – Milk – Fatty acids – Amino acids.

Effet du remplacement partiel du concentré par des blocs alimentaires contenant des sous-produits de la tomate issus de cultures sous serre, sur la production de méthane et la production et la composition du lait chez des chèvres

Resumé. Ce travail avait pour objectif l'étude, chez des chèvres laitières, du remplacement de 35% de la fraction concentrée de la ration par des blocs alimentaires contenant des sous-produits de tomate sur les émissions de CH₄, la production laitière et la composition du lait (acides gras et acides aminés). Huit chèvres de la race Granadina (39,6 ± 1,89 kg poids vif) en milieu de troisième lactation ont participé à deux essais selon un schéma en cross-over. Dans chaque essai, 4 animaux tirés au hasard ont reçu 1,0 kg de foin de luzerne (AH), plus 1 kg de concentré (régime AC), alors que les 4 autres ont reçu 1,0 kg AH plus 0,65 kg de concentrés et des blocs alimentaires contenant des tomates offerts en quantité *ad libitum* (régime ACB). Le régime ACB a entraîné une réduction de 38,3% (P = 0,001) des émissions de CH₄, et une augmentation dans le lait (P ≤ 0,048) des proportions des acides linoléique et linoléique et des acides gras polyinsaturés totaux. Le profil en acides aminés et leur concentration n'ont pas été affectés par le régime, mais la proportion de la phénylalanine a été plus faible (P = 0,039) dans le lait des animaux recevant le régime ACB par rapport au témoin. En conclusion, le régime ACB distribué aux chèvres favorise la production d'un lait avec un profil en acides gras de meilleure valeur nutritionnelle tout en réduisant le coût de l'alimentation et les impacts environnementaux négatifs de la production caprine liés aux émissions de méthane.

Mots-clés. Chèvre – Blocs alimentaires – Tomate – Méthane – Lait – Acides gras – Acides aminés.

I – Introduction

Ruminant production in the Mediterranean area is limited by the poor quality and scarcity of pasture. Thus concentrates based on cereals are frequently used, but increase in cereal prices has driven the attention of ruminant nutritionists toward local alternatives (Ben Salem and Znaidi, 2008). Greenhouse horticulture is very important in the area, producing large amount of wastes, which could be an alternative to cereals for ruminants. Feed blocks manufacturing allows the inclusion of high-moisture wastes in animal feeding (Ben Salem and Nefzaoui, 2003). Milk fatty acid profile can be manipulated by the inclusion of some agro-industrial by-products in the diet depending on their energy value, fatty acid composition and fibre content (Vasta *et al.*, 2008). Nevertheless, few studies have investigated the use of by-products in dairy goats, analysing milk fatty acid composition (Molina-Alcaide *et al.*, 2010a; Modaresi *et al.*, 2011). As for conventional protein sources, the success of the utilization of alternative protein sources in dairy goat feeding depends on the ability to formulate diets balanced in essential amino acids (Vasta *et al.*, 2008). Moreover the presence of plant secondary and other unknown compounds in greenhouse wastes could modify rumen fermentation and thus, methane emission (Patra and Saxena, 2010). The aim of the present experiment was to study, in dairy goats, the effect of replacing 35% of concentrate in the diet with feed blocks including wastes from tomato fruits on CH₄ emission and on milk yield and fatty acid and amino acid profiles.

II – Materials and methods

Eight Granadina goats (39.6±1.89 kg BW) in the middle of the third lactation were used, and 2 periods were carried out following a crossover design. In each trial, 4 animals randomly received 1.0 kg of alfalfa hay (AH) plus 1 kg of concentrate (diet AC) and the other 4 received 1.0 kg AH plus 0.65 kg of concentrate plus feed blocks (B) including greenhouse wastes of tomato (diet ACB) with 8 replications per diet at the end of the trials. Animals were individually kept in boxes with free access to food and water. Ingredient composition of concentrate and tomato FB is shown in Table 1. The amounts of AH and concentrate supplied to the animals fed diet AC were sufficient to allow daily milk production of up to 2 kg per goat (Aguilera *et al.*, 1990). The feed block was supplied *ad libitum*, resulting in average intakes of 208±31.8 g of DM/animal/d for ACB diet. Each trial consisted of 25-d for adaptation and 8-d for sampling. Individual intakes of diet ingredients were registered through the whole trial. Goats were hand-milked once a day before feeding and milk yield recorded and milk density was measured and aliquots were stored at -30°C without preservatives until analyzed. The last 3 days of each trial animals were individually placed into square polycarbonate chambers (1.8×1.8×1.5 m) to measure CH₄ emissions. Ground (1-mm) samples of ingredients were analyzed for dry matter (DM), organic matter (OM), ether extract (EE) and total N (Table 2) according to the AOAC (2005). The NDF (neutral detergent fibre) and ADF (acid detergent fibre) were analyzed according to van Soest *et al.* (1991) using an ANKOM Model 220 Fiber Analyzer (Macedon, NY, USA) with α -amylase for NDF analysis in concentrate samples and both NDF and ADF contents referred to ash-free weight. The ADL (acid detergent lignin) was determined by solubilisation of cellulose with 72% sulphuric acid. The energy content was analysed using an oxygen bomb calorimeter (PARR 1356, Biometer). Total N content in feedstuffs and milk was determined as described by (Molina-Alcaide *et al.*, 2010a). Extraction of total fatty acids in feedstuffs was based on the method of Folch *et al.* (1957), with modifications (Devillard *et al.*, 2006). Total fatty acids in milk were extracted as described by Toral *et al.* (2011). The amino acid N (AA-N) content in samples of feeds and milk was determined by HPLC using the Waters® (Waters Corporation, Mildford, MA, USA) following the procedures described by Molina-Alcaide *et al.* (2010b). Methane emission was calculated from CH₄ concentration analysed using a gas analyzer ADM MGA3000 (Spurling Works, Herts, UK) and airflows into and out of each chamber. Data were analyzed by GLM (general linear models) using repeated measures of ANOVA. Diet was considered as a fix effect, and trial and animal as random effects. When a significant effect of diet was found, post hoc comparison of means was made using the LSD

test. Differences were considered significant at $P < 0.05$ and $P < 0.10$ values were declared as trends and discussed.

Table 1. Ingredients composition of concentrate and feed block (g/kg fresh matter)

Ingredient	Concentrate	Feed block
Wheat shorts	350	—
Corn grain	50	—
Barley	160	—
Tomato waste	—	585
Sunflower meal	115.5	36
Soybean hulls	90	—
Corn middlings	90	—
Soybean meal	90	—
Wheat straw	—	221
Beet molasses	—	100
Fatty acid salts	4.5	—
Quicklime	22	27
NaCl	3.0	16
Vitamin-mineral mixture [†]	25	3.3
Urea	—	11.7

[†]Formulated (per kg) with NaCl, 277 g; ashes from the two-stage dried olive cake combustion, 270 g; $(\text{PO}_4)_2\text{H}_4\text{Ca}$, 250 g; MgSO_4 , 200 g; CuO, 184 mg; I, 25 mg; CoO, 8.5 mg; Se, 4 mg; ZnO, 2.28 mg; and 83,500 and 16,700 IU of vitamins A and D, respectively.

III – Results and discussion

The composition of diets in ingredients is shown in Table 1. The chemical composition of diet components is shown in Table 2.

Table 2. Chemical composition (g/kg DM) and gross energy (GE) of alfalfa hay, concentrate and feed blocks (n=3)

Item	Alfalfa hay	Concentrate	Feed block
DM, g/kg fresh matter	906	926	907
OM	881	893	814
CP	212	170	165
NDF	417	338	466
ADF	251	143	273
ADL	59	25	44.7
Ether extract	13.8	34.1	7.29
GE, MJ/kg DM	18.2	18.2	16.0

Diet did not affect total dry matter intake or animals live weight (data not shown). The ACB diet resulted in a 38.3% reduction ($P = 0.001$) of CH_4 emission (Table 3). It has been reported that plant secondary compounds, like tannins or saponins, could modify rumen fermentation,

inhibiting enteric methanogenesis (Guo *et al.*, 2008; Patra and Saxena, 2010). The presence of these compounds in tomato byproducts may have contributed to the methane reduction by (i) decreasing OM fermentation in the rumen and increasing OM fermentation in the intestine; (ii) diverting hydrogen away from CH₄ production; and (iii) and inhibition of microbial activity or optimization of rumen fermentation, decreasing methane emission per unit of OM digested. However, the association between the antimethanogenic effect of plant secondary compounds and their molecular structure and weight together with chemical composition of diets, should be taken into account (Newbold *et al.*, 2004).

Table 3. Methane emissions, milk production and fatty acid composition (g/100 g of identified FA), and milk recovery rate (g of FA excreted in milk/g of FA intake) of selected fatty acids of goats fed with different experimental diets

Item	Diet [†]		SEM	P-value
	AC	ACB		
CH ₄ emission, L/kg DMI	28.2 ^b	17.4 ^a	0.819	0.001
Milk yield, ml/d	997	922	94.16	0.372
Total fat, g/L milk	56.2	56.8	3.589	0.846
Total saturated FA	74.7	73.4	0.650	0.319
Total Monounsaturated FA	19.7	20.3	0.672	0.565
Poly unsaturated FA				
<i>cis</i> -9, <i>cis</i> -12, 18:2	1.94 ^a	2.17 ^b	0.069	0.048
<i>cis</i> -9, <i>trans</i> -11, CLA	0.57	0.62	0.061	0.487
<i>trans</i> -11, <i>cis</i> -15 18:2	0.037 ^a	0.053 ^b	0.006	0.033
<i>cis</i> -6, <i>cis</i> -9, <i>cis</i> -12, 18:3	0.017	0.025	0.002	0.503
<i>cis</i> -9, <i>cis</i> -12, <i>cis</i> -15, 18:3	0.39 ^a	0.54 ^b	0.035	0.028
20:3 n-6	0.020	0.027	0.005	0.244
20:4 n-6	0.20	0.23	0.012	0.082
Total Polyunsaturated FA	3.34 ^a	3.83 ^b	0.149	0.012
FA recovery rate				
<i>cis</i> -9, <i>cis</i> -12, <i>cis</i> -15, 18:3	0.090	0.12	0.011	0.063
<i>cis</i> -9, <i>cis</i> -12, 18:2	0.13 ^a	0.17 ^b	0.020	0.021
³ <i>cis</i> -9 18:1	1.16 ^a	1.48 ^b	0.194	0.012
18:0	1.57	1.63	0.204	0.569

[†]AC = Alfalfa hay and concentrate (1:1); ACB = alfalfa hay, concentrate (1:0.65) and tomato feed block.

*About 2.4 of non identified fatty acids.

Milk yield, as well as fat and protein concentrations, were not affected ($P \leq 0.907$) by the inclusion of feed block in the diet, which may be due to the similar energy concentration and rumen protein degradability of both diets. Dietary intakes of linoleic (LA; *cis*-9, *cis*-12, 18:2) and linolenic (LNA; *cis*-9, *cis*-12, *cis*-15, 18:3) acids (data not shown) showed that goats feeding the diet including tomato fed block consumed less unsaturated fatty acids than those under the control diet. However, milk from goats fed the diet including block showed higher content of LA ($P=0.048$), LNA ($P=0.028$) and total polyunsaturated fatty acids (PUFA) ($P=0.012$); a concomitant tendency ($P=0.054$) to decrease 18:0 (SA) concentration (11%) was also observed when tomato feed block was fed. Regarding *cis*-9, *trans*-11CLA concentrations, milk fat of goats on tomato feed block diet remained unchanged ($P=0.487$) in comparison to milk from animals fed diet AC. The higher ($P<0.033$) accumulation of *trans*-11, *cis*-15, 18:2 in milk from goats fed diet including tomato fed block would suggest an incomplete biohydrogenation of LNA. In addition to the fatty acid supply, other factors such as energy supply, the proportion of fibre or

concentrate as well as the presence of plant secondary compounds (Leiber *et al.*, 2005; Vasta *et al.*, 2008) should be considered when dietary effects on milk fatty acid profile are assessed. Since the fatty acid supply was lower in the ACB diet than in the AC diet, and taking into account that energy and fiber intakes were similar for goats fed both diets (data not shown), it might be that the observed effect in milk fatty acid composition could have been due to the administration of the ingredients in the feed block. The synchronous and fractionated supply of nutrients allowed when using feed blocks in ruminants feeding (Ben Salem and Nefzaoui, 2003) may have been associated to a better fatty acid absorption in the small intestine. Additionally, it could be speculated that during the manufacture of the block tomato seeds were partially crushed so one fraction of fatty acids may have been accessible to biohydrogenating bacteria leading to accumulation of biohydrogenation intermediates, whereas another fraction could have been still protected and therefore higher amounts of PUFA would have been available in the small intestine for absorption. Fatty acid recovery rates were higher for LA and oleic acid (OA; *cis*-9, 18:1) when feeding diet ACB which would support the previous speculation. This finding was in agreement with those of Abbeddou *et al.* (2011) and Khiaosa-Ard *et al.* (2010) who showed increased linoleic acid recovery rates in milk following decreased linoleic acid intakes. Also, other components of the tomato by-products might have been involved in the observed effect; for example, tannins and saponins have been shown to be of potential usefulness in controlling biohydrogenation (reviewed by Lourenço *et al.*, 2010). Overall, milk of goats fed tomato block diet could be considered of healthier fatty acid composition as increased amounts of LA and LNA (1.12 and 1.38 fold, respectively) in the milk would be important due to their beneficial effects in the prevention of cardiovascular diseases and hypertension in humans.

Total N and amino acid profile of milk (Table 4) were unaffected by diet ($P \geq 0.215$) with the exception of phenylalanine that was lower ($P = 0.039$) in the milk of goats fed diet ACB compared to AC. Taking into account the relative proportions of cereals in concentrate and in feed blocks, their average intakes in the current study, and the cost of cereals and milk (MARM, 2012), a reduction in feeding cost of 22% may be achieved with diets containing feed blocks compared to the control diet.

IV – Conclusions

Feed blocks based on tomato fruits wastes could replace 35% of the concentrate in dairy goats diet without detrimental effects on milk production and fatty acid or amino acid composition. Overall, diet including tomato feed blocks promoted the production of milk with healthier fatty acid profile and reduced feeding cost and the environmental damage associated with by-products accumulation and CH₄ emissions compared with a conventional diet.

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Table 4. Total N and amino acids profile of goats milk fed with different experimental diets.

Item	Diet [†]		SEM	P-value
	AC	ACB		
Total N, mg/g DM	40.6	40.4	1.21	0.907
Total amino acids, g/L milk	5.10	4.91	0.234	0.451
Total amino acids ^{††} , g/100g total N	84.0	84.6	2.78	0.900
AA, g/100 g total AA				
Aspartic acid	6.38	6.69	0.254	0.244
Glutamic acid	18.5	18.6	0.331	0.867
Serine	4.47	4.64	0.091	0.332
Glycine	1.72	1.75	0.042	0.667
Histidine	3.57	3.59	0.044	0.753
Arginine	2.50	2.79	0.113	0.237
Threonine	4.62	4.63	0.096	0.962
Alanine	3.10	3.15	0.131	0.687
Proline	8.20	8.28	0.189	0.791
Tyrosine	4.61	4.70	0.168	0.528
Valine	5.54	5.49	0.141	0.843
Methionine	8.15	7.65	0.571	0.744
Cysteine	4.14	4.23	0.419	0.902
Isoleucine	4.38	4.31	0.041	0.460
Leucine	7.53	7.43	0.123	0.592
Phenylalanine	4.28 ^b	4.12 ^a	0.073	0.039
Lysine	8.31	7.94	0.104	0.215
EAA ^{†††}	48.9	48.0	0.489	0.290
NEAA ^{††††}	51.1	52.0	0.480	0.285

[†]AC = Alfalfa hay and concentrate (1:1); ACB = alfalfa hay, concentrate (1:0.65) and tomato feed block.

^{††}Without tryptophane. ^{†††}EAA: threonine, arginine, valine, methionine, isoleucine, leucine, phenylalanine, lysine and histidine. ^{††††}NEAA: alanine, aspartic acid, glutamic acid, glycine, proline, serine, tyrosine and cysteine.

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