

Blood Biochemical Profile and Rumen Fermentation Pattern of Goats Fed Leaf Meal Mixture or Conventional Cakes as Dietary Protein Supplements

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ABSTRACT : The expediency of replacing cost prohibitive and often inaccessible traditional protein supplements prompted the current investigation of blood biochemical profile and rumen fermentation pattern in local female goats (12) and rumen fistulated bucks (3), respectively fed supplements containing either a leaf meal mixture (LMTM) of *Leucaena leucocephala*-*Morus alba*-*Tectona grandis* (2:1:1) or traditional protein supplements groundnut cake (GNC) or soybean meal (SBM) and wheat straw as basal diet. The periodic monitoring of hematological parameters was carried out in female goats at 0, 30, 60 and 90 days post feeding. Rumen environment was studied in bucks in a 3×3 switch over design. Rumen liquor was collected at 0, 2, 4, 6 and 8 h post feeding after 4 weeks of feeding. The goats fed on LMTM or GNC had similar dry matter intake (g/kg W^{0.75}), which was significantly (p<0.05) higher than SBM. Except for packed cell volume (PCV), none of the blood biochemical constituents (Hemoglobin, serum glucose, total protein, serum albumin (A) and globulin(G), A:G ratio, alkaline phosphatase, transaminases) varied significantly due to replacement of 50% dietary protein by LMTM throughout the experiment. GNC group had significantly higher level of PCV than other treatments. However, the level of serum total protein (p<0.01) tended to increase from 60th day onwards irrespective of dietary treatments. The average rumen pH was significantly higher (p<0.001) on SBM followed by LMTM and GNC, respectively. Total volatile fatty acid (TVFA) production was comparable in goats given LMTM or GNC supplements, the corresponding values were significantly different (p<0.001) when compared with SBM. The ammonical-N, total-N and TCA-precipitable-N (mg/100 ml SRL) did not differ significantly among dietary treatments. It may be concluded that supplementing wheat straw with LMTM based concentrate had no adverse effect on voluntary intake, blood biochemical profile and rumen fermentation pattern of the goats. (*Asian-Aust. J. Anim. Sci.* 2002. Vol 15, No. 5 : 665-670)

Key Words : Goat, Leafmeal Mixture, Protein Supplement, Rumen Fermentation, Blood Traits

INTRODUCTION

The goats are under pressure to switch over from traditional free ranging system to stall feeding due to increasing population and land shortage. Therefore, ways and means have to be found to use the poor quality agricultural by products better and more than the current extent for small ruminant production. A large quantity of cereal straws is available in our country, which together with other crop residues have to be classified as poor quality feeds because of their high fibre content, low protein and unbalanced mineral composition (Sundstol and Owen, 1984). Farmers traditionally used protein supplements (oil cakes, bran and cereals) to improve nutritive value of fibrous crop residues, but the high cost and poor accessibility of these protein supplements is prohibitive. Trees and shrubs have a great value as dietary protein supplements (Devendra, 1990). The use of multipurpose tree species as a supplement or a component in conventional concentrate has been shown to improve utilization of cereal straws (Narayan Dutta et al., 1999). The

importance of *Leucaena leucocephala*, *Morus alba* and *Tectona grandis* as multipurpose trees is increasingly felt in various agro-forestry and production systems. However, wide spread use of these tree fodders has been restricted due to the presence of anti nutritional factors like mimosine, tannins or low palatability to livestock. Farmers usually minimize and overcome these problems by feeding mixture of tree leaves with or without sundrying (Lowry, 1990). Supporting evidence for nutritional feasibility and benefits of dietary incorporation of *Leucaena leucocephala*, *Morus alba* and *Tectona grandis* leaf meal mixture up to 20% has been obtained from our studies with goats (Anbarasu et al., 2001). Present investigation was undertaken to investigate the effect of supplementing wheat straw with leaf meal mixture as a protein supplement substituting conventional oil cakes on blood biochemical profile and rumen fermentation pattern of goats.

MATERIALS AND METHODS

Animal management and experimental feeding

The experiment was conducted during January-April, 2000 at Animal Nutrition Division of Indian Veterinary Research Institute to study the efficacy of leaf meal mixture in the ration of goats. Twelve female goats, about 9 months old (Avg. B.wt. 11.4±0.6 kg) were allotted to three dietary

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Received October 8, 2001; Accepted January 7, 2002

treatments (4 goats in each group) in a complete randomized block design. They were offered a basal diet of wheat straw *ad libitum* and either of the three concentrate mixtures computed as indicated in table 1. The control group concentrate (GNC) was formulated by using deoiled groundnut cake as the main protein supplement while deoiled soybean meal and leaf meal mixture formed part of concentrate mixtures SBM and LMTM, respectively by replacing about 50% CP of GNC concentrate. The leaf meal mixture component of LMTM concentrate contained sundried ground leaves (% DM) of *Leucaena leucocephala* 50, *Morus alba* 25 and *Tectona grandis* 25. The leaves were individually sundried in one lot in the months of June-July and ground in an electric grinder before mixing in the ratio of 2:1:1. The goats were offered respective concentrates in a single meal at 09:00 h in the morning followed by wheat straw *ad-libitum* to meet their requirement for maintenance and growth (25 g/d) as per NRC (1981). Leftovers of wheat straw were weighed 24 h post feeding to ascertain daily feed consumption. The experiment was carried out for 15 weeks. Daily DM intake and fortnightly live weight of all the goats were recorded in the morning prior to feeding. Proximate and cell wall composition of offered feed and leftovers were determined by the method of AOAC (1995) and Goering and VanSoeset (1970), respectively. Total tannins in the LMTM was determined by oxidation with acid permanganate on a volumetric basis (AOAC, 1995).

Table 1. Ingredients and chemical composition (%DM) of concentrates

Formula/attributes	Experimental diets			
	GNC	SBM	LMTM	Wheat straw
Ingredients				
Leaf meal mixture	-	-	45	
Deoiled groundnut cake	35	-	17.5	
Deoiled soybean meal	-	26	-	
Maize	21	21	21	
Wheat bran	42	51	14.5	
Mineral mixture	1	1	1	
Common salt	1	1	1	
Chemical composition				
Organic matter	90.44	91.58	88.10	92.47
Crude protein	22.99	24.56	24.21	3.12
Ether extract	2.79	3.33	4.35	1.03
Total ash	9.56	8.42	11.90	7.53
NDF	28.99	27.56	29.05	79.29
ADF	9.36	7.12	12.11	47.41
Total tannins	-	-	3.64	-
Mimosine	-	-	6.12	-

GNC: groundnut cake, SBM: soybean meal and LMTM: leafmeal based supplements.

The mimosine content of *Leucaena leucocephala* and LMTM concentrate was measured by the method of Megarrity (1978).

Blood collection and blood-biochemical profile

Blood samples were collected in the morning before feeding at 0, 30, 60 and 90 days post feeding by jugular vein puncture. Serum was separated from about 8 ml of whole blood collected from each animal and stored at -20°C. Another 2 ml blood sample was collected in tubes containing ethylene diaminetetraacetate at 1 mg/ml blood, for hematological parameters. Hemoglobin and packed cell volume (PCV) were estimated in whole blood immediately after the collection of blood by acid haematin method (Benjamin, 1985) and Wintrobe's tube (Hawk, 1965), respectively. Serum glucose concentration was determined colorimetry (Hultmann, 1959). The total serum protein and albumin (A) content of serum were measured as per Wotton (1964) and Doumas et al. (1971). Globulin (G) values were obtained by subtracting the value of albumin from total protein and A:G ratio was calculated. The activity of serum glutamate-oxaloacetate transaminase (SGOT) and serum glutamate-pyruvate transaminase (SGPT) (Reitman and Frankel, 1957) and alkaline phosphatase (King and Armstrong, 1934) were estimated as per standard colorimetric methods using reagent kits supplied by M/s Qualigens Fine Chemicals (A division of Glaxo India Ltd.) with the help of spectrophotometer (Spectronic 20).

Rumen fermentation studies

Three rumen fistulated bucks (Avg BW 26.0±0.5 kg) about 3 years old, were used in a 3×3 switch over design. The animals were fed a basal diet of wheat straw *ad-libitum* and supplemented with either GNC, SBM or LMTM concentrate to meet their maintenance requirement (NRC, 1981). Drinking water was offered twice daily at 9:30 h and 14:30 h. After 4 weeks of preliminary feeding during each period, rumen liquor was collected at 0, 2, 4, 6 and 8 h post feeding for three consecutive days. The rumen pH was recorded with a digital pH meter. The strained rumen liquor (SRL) was preserved in deep freeze with few drops of saturated solution of mercuric chloride for further analysis. The ammonical nitrogen (NH₃-N), trichloroacetic acid precipitable nitrogen (TCA-ppt-N), total nitrogen and total volatile fatty acids (TVFA) were determined (Conway, 1957; AOAC, 1995; Annison, 1954).

The data obtained were subjected to analysis of variance in a completely randomized block design and for rumen metabolites in latin square design ignoring the period effect. Treatment means were ranked using Duncan's multiple range test (Snedecor and Cochran, 1989).

RESULTS AND DISCUSSION

Voluntary intake

Daily dry-matter intake (DMI; $\text{g/kg W}^{0.75}$) of goats was significantly ($p < 0.05$) higher for LMTM group as compared to SBM. However, the feed intake of goats on LMTM was similar to GNC group. Intake of various concentrate mixtures by goats was similar irrespective of protein supplement (figure 1). Therefore, the difference observed in total DMI of goats among different dietary groups was due to variation in intake of wheat straw component of the diet. The factors responsible for variable effects of dietary supplements on wheat straw intake may include difference in rate of passage of digesta, packing density, dietary concentration of ruminally degradable protein, availability of branch chain fatty acids, rapidly fermentable β -glucose and some hitherto unknown microbial growth promoters especially in leguminous forages (Goodchild and McMeniman, 1994; Khandaker et al., 1998; Sharma et al., 1998). Interestingly, despite bulky nature of LMTM concentrate, it maintained or boosted straw intake of goats at par with conventional vegetable protein concentrates GNC or SBM, respectively.

Blood-biochemical profile

The goats remained apparently in good health throughout the experiment irrespective of dietary supplement. The initial (11.37 ± 0.6 kg) and final (13.64 ± 0.7 kg) body weights of goats were not influenced by nature of protein supplement and thus accounted for the non-significant differences in the net live weight gain (2.27 ± 0.2 kg). These results are in agreement with the reports of other workers (Gupta et al., 1991; Tewatia et al., 1997; Mahanta et al., 1999) indicating comparable body condition in goats on tree leaves or oil cakes.

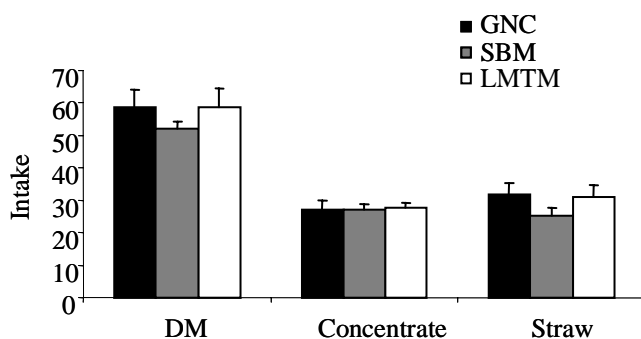


Figure 1. Feed intake ($\text{g/kg BW}^{0.75}$) of goats (GNC: groundnut cake, SBM: soybean meal, and LMTM: leafmeal based supplements).

The levels of hemoglobin and packed cell volume ranged from 10.50 to 11.62 (g %) for hemoglobin and 30.25 to 34.25 (%) for PCV, which were within the normal range (8-12 g % Hb and 24-48% PCV) as reported by Kaneko (1989) and Benjamin (1985). This suggests that the general health of goats given three different diets remained normal and similar. However, the significantly higher mean PCV values of goats given GNC treatment may be due to the significantly ($p < 0.05$) higher level of PCV initially observed in this group (table 2) which remained unaltered throughout the experiment. The serum glucose level (45.42 to 49.08 mg dl^{-1}) was not influenced by the dietary treatment or exposure time (Period) of the diet. Our finding contradicts with Kailash (1991), who observed a significant decrease in blood glucose level of goats given diets containing 50% CP replaced by *Leucaena*, suggesting hypoglycemic effect of mimosine (Akbar and Gupta, 1985). However, the use of low mimosine variety of *Leucaena* in the study resulted in very low intake of mimosine (0.03 $\text{g/kg W}^{0.75}$) which was too low to cause any deleterious effect (Hammond, 1995).

The level of alkaline phosphatase (IUL^{-1}) activity ranged from 129.27 to 177.96 during the study, with no evidence of any effect of dietary treatment or period of measurement. The observed level of alkaline phosphatase was within the normal range as reported by Kaneko (1989). This suggests that feeding of LMTM to goats had no adverse impact on bone and liver though it contained moderate amount of antinutritional factors such as tannins. The serum glutamate oxaloacetate and serum glutamate pyruvate transaminase activities remained within normal range (table 2) throughout this study in goats irrespective of dietary supplement. This is also indicative of no adverse effect of leafmeal or conventional cake based concentrate on vital organs like liver or muscles (Evans, 1988).

The level of total serum protein ranged from 6.53 to 7.25 g/dl^{-1} in all groups during the different feeding period. The observed range of total serum protein in experimental goats was within normal range of 6 to 7.5 g/dl^{-1} (Kaneko, 1989) irrespective of dietary treatment (table 3). However, the serum protein level tended to increase ($p < 0.01$) from 60th day of experiment. Rajendran (1999) also reported an increasing trend of serum protein in goats fed diets with 50% CP replaced by *Leucaena* leafmeal. Similarly, feeding of SBM at different levels also increased the total protein in serum (Lu et al., 1990). Serum albumin, globulin and A:G ratio ranged from 2.90 to 3.74, 2.79 to 4.13 and 0.81 to 1.29, respectively and showed no significant difference among different dietary supplements. Though the albumin level was significantly ($p < 0.05$) higher at 30 days and the globulin was significantly ($p < 0.05$) lower

Table 2. Effect of supplementation on hemoglobin (g dl⁻¹), packed cell volume (percent), serum glucose (mg dl⁻¹), serum enzyme level (IU/ml)

Attributes	Experimental diets			SEM
	GNC	SBM	LMTM	
Hemoglobin	11.56	11.04	10.82	0.20
Packed cell volume	34.50 ^b	31.10 ^a	31.63 ^a	0.85
Glucose	47.01	48.11	46.87	0.56
Alkaline phosphatase	145.76	157.69	159.87	8.85
SGOT	56.65	57.36	53.84	1.45
SGPT	49.38	49.50	50.20	1.70

^{a,b} Mean values bearing different superscripts in a row differ significantly ($p < 0.05$).

GNC: groundnut cake, SBM: soybean meal and LMTM: leafmeal based supplements.

SGOT: serum glutamate-oxaloacetate transaminase, SGPT: serum glutamate-pyruvate transaminase.

Table 3. Effect of supplementation on total serum protein (g dl⁻¹), serum albumin (g dl⁻¹), serum globulin (g dl⁻¹) and albumin:globulin ratio

Attributes	Experimental diets			SEM
	GNC	SBM	LMTM	
Total serum protein	6.74	6.77	6.95	0.05
Serum albumin	3.30	3.43	3.36	0.11
Serum globulin	3.38	3.27	3.68	0.12
Albumin:globulin ratio	1.10	1.00	1.15	0.06

^{a,b} Mean values bearing different superscripts in a row differ significantly ($p < 0.05$).

GNC: groundnut cake, SBM: soybean meal and LMTM: leafmeal based supplements.

at the initiation of the trial in goats, the A:G ratio remained unaltered in all the animals throughout the feeding period. The serum protein profile (albumin, globulin, A:G ratio) remained within the normal range prescribed for healthy goats (Kaneko, 1989).

Rumen fermentation

Average rumen pH (table 4) was significantly ($p < 0.01$) lower on LMTM as compared to SBM. However, the rumen pH of goats on GNC was significantly lower to LMTM and SBM. The rumen pH steadily declined from 0 h to 8 h post feeding irrespective of dietary treatment though the reduction was more pronounced ($p < 0.01$) after 4 h post feeding. The difference in rumen pH is an indication of varying buffering capacity and rate of degradation of different protein supplements (Narayan Dutta and Singh 1994; Reddy et al., 1989). The TVFA concentration (m.moles/l) was comparable in goats given LMTM or GNC supplements, the corresponding values were significantly higher ($p < 0.01$) when compared with SBM. The lower production of TVFA on SBM group could be due to lower solubility of nitrogen and reduced availability of substrates

viz. amino acids for production of VFAs. Present results are in agreement with earlier reports (Grieve et al., 1980; Sharma et al., 1972). TVFA concentration peaked at 4 h post feeding in the animals and declined thereafter upto 8 h post feeding. Results obtained were in agreement with the findings of Sinha et al. (1974) and Singh et al. (1983). Ruminant NH₃-N (mg/100 ml SRL) concentration was similar among three dietary treatments and peaked at 2 h post feeding. Similarly, the average concentration (mg/100 ml SRL) of total-N and TCA-ppt-N was comparable in animals under three dietary supplements. It is understandable because goats were fed on isonitrogenous diets. The total-N concentration was significantly ($p < 0.01$) higher from 2 to 4 h post feeding and thereafter declined till 8 h post feeding. The TCA-ppt-N peaked significantly ($p < 0.01$) from 2 to 6 h post feeding and declined thereafter. The results obtained were in conformity with those reported by Narayan Dutta and Singh (1994) and Reddy and Reddy (1986). No significant interaction was evident between treatments and periods for any of the rumen metabolites.

Overall, it can be confirmed clinically that goats fed LMTM supplement do not have a biochemically determinable disadvantage to their counterparts fed groundnut cake or soybean meal as a vegetable protein source. The *Leucaena leucocephala*-*Morus alba*-*Tectona grandis* based leaf meal mixture could contribute 20% of total ration of goats without any adverse effect on their performance.

ACKNOWLEDGEMENT

This study was financially supported by funds provided by Indian Council of Agricultural Research (AP-Cessfund), NewDelhi, India.

Table 4. Effect of different protein supplements on rumen environment

Variable	Treatment	Sampling time (h)					Mean±SEM
		0	2	4	6	8	
pH							
	GNC	6.72	6.50	6.42	6.46	6.33	6.48 ^b ±0.04
	SBM	7.21	7.01	7.07	7.01	7.03	7.06 ^c ±0.05
	LMTM	6.89	6.83	6.58	6.58	6.54	6.68 ^a ±0.05
	Mean±SEM	6.94 ^b ±0.07	6.78 ^{ab} ±0.08	6.69 ^a ±0.08	6.68 ^a ±0.079	6.63 ^a ±0.08	
NH ₃ -N (mg/100 ml SRL)							
	SBM	11.50	15.50	15.50	13.50	12.50	13.70±0.51
	GNC	13.00	18.25	17.50	13.40	11.75	14.79±0.59
	LMTM	10.00	19.33	15.66	13.66	10.66	13.86±0.75
	Mean±SEM	11.50 ^a ±0.53	17.69 ^b ±0.74	16.22 ^b ±0.61	13.54 ^a ±0.36	11.64 ^a ±0.33	
Total-N (mg/100 ml SRL)							
	GNC	103.75	157.50	163.75	136.25	108.25	133.90±4.80
	SBM	105.00	170.00	162.50	135.00	116.25	137.75±4.86
	LMTM	162.50	163.33	165.83	152.00	110.00	140.83±5.44
	Mean±SEM	103.75 ^a ±2.23	163.61 ^c ±4.95	164.03 ^c ±4.25	141.08 ^b ±4.02	111.1 ^a ±4.95	
TCA-ppt-N (mg/100 ml SRL)							
	GNC	81.25	105.00	115.00	97.00	85.75	96.83±2.36
	SBM	81.00	108.75	112.50	100.75	88.75	98.55±2.76
	LMTM	77.50	105.00	116.66	119.16	79.66	99.83±3.71
	Mean±SEM	79.97 ^a ±1.96	106.58 ^b ±3.17	114.72 ^b ±3.03	105.64 ^b ±3.58	84.72 ^a ±3.52	
TVFA (m.moles/l)							
	GNC	69.16	91.66	90.82	86.60	77.46	83.15±2.1
	SBM	54.16	76.24	80.81	79.15	74.58	78.99±1.8
	LMTM	76.65	84.99	91.65	90.53	83.60	85.49±1.9
	Mean±SEM	66.66 ^a ±2.89	84.30 ^b ±3.87	87.76 ^b ±3.01	85.44 ^b ±2.75	78.55 ^b ±3.10	

^{a,b,c} Mean values bearing different superscripts in a row and column differ significantly (p<0.001).

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