Pak. J. Bot., 43(1): 397-404, 2011.

CHEMICAL COMPOSITION, MINERAL PROFILE AND *IN SITU* DIGESTION KINETICS OF FODDER LEAVES OF FOUR NATIVE TREES

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Abstract

This study explored the nutritive value of *Morus alba, Acacia nilotica, Syzygium cumuni* and *Ziziphus jujuba* leaves. Chemical analyses revealed that dry matter (DM) ranged from 25% to 47% in *M. alba* and *S. cumunii*, organic matter was higher (94%) in *S. cumuni* and *Z. jujuba*. *M. alba* had higher (23%) crude protein, whereas, neutral detergent fiber (NDF) was greater in *Z. jujuba* (32%). Acid detergent fiber was higher in *S. cumuni* (23%), while, acid detergent lignin was greater (7%) in *M. alba* and *S. cumuni*. Hemicellulose (15%) and ash content (10%) were higher in *Z. jujuba* and *M. Alba*, respectively. Metabolizable energy was higher (10.5 MJ/kg) in *M. alba* than the other species. Among minerals, Ca and K were highest in *A. nilotica*, P in *M. alba* and Mg and Na in *Z. jujuba*. The concentration of total phenolics, tannins, alkaloids and saponin was within safe range. *In situ* DM digestibility was higher (90.2%) for *M. alba*, DM lag time was shorter (0.63 h) for *A. nilotica*, and rate of DM disappearance was lowest (5.34% per h) for *S. cumuni*. Extent of DM digestion (98.26%) and NDF digestibility (84.10%) were higher for *M. alba*. Shorter NDF lag time (0.71 h) and higher rate of NDF disappearance were evident for *A. nilotica*, but extent of NDF digestion was higher (96.80%) for *M. alba*. Based on chemical composition and *in situ* digestion kinetics, *M. alba* leaves proved the best supplement followed by *A. nilotica, Z. jujuba* and *S. cumuni* for ruminants.

Introduction

Availability of feed containing imbalanced chemical composition and metabolizable energy (ME) is major handicap in ruminant production the world over (Niderkorn & Baumonta, 2009). There is a great need to feed the ruminant animals with balanced feed to improve the production of meat and milk. Several tree species could be effective sources of providing fodder nutrition during normal as well as scarcity periods (Reddy, 2006). Although there are many advantages of the forages and leguminous crops over tree crops, the leaves of certain trees can be as nutritious as those of fodder legumes (Soliva *et al*., 2005; Anon., 2006).

Area under fodder production is continuously reducing due to competition with cash crops. The ever increasing demand of cereal grains for human consumption coupled with reduction in land for fodder cultivation is decreasing the nutrient supply to ruminants. Thus, there is great need to explore alternate feed resources which do not compete with human feed (Raghuvansi *et al*., 2007). Fodder tree leaves are alternative feed source for ruminant (Malik *et al*., 1967) and can help to minimize the wide gap between availability and supply of nutrients, and improve the animal growth and productivity. Fodder tree leaves were found to be rich in protein, soluble carbohydrates, minerals and vitamins, and showed great potential as an alternate feed resource (Baumer, 1992; Bakshi & Wadhwa, 2007). Use of tree leaves in ruminant enhances microbial growth and digestion (Singh *et al*., 1982; Bonsi *et al*., 1995). Moreover, fodder tree leaves are very relish to small ruminants especially goats.

Table 1. Name of trees.

Botanical name	Local name	Family
Morus alba	Toot, mulberry	Moraceae
Acacia nilotica	Kikar	Mimosaceae
Syzygium cumuni	Jaman	Myrtaceae
Ziziphus jujuba	Beri	Rhamnaceae

In view of little information on the nutritive value of fodder tree leaves especially digestion kinetics and ME availability in ruminants, the present study was planned to figure out the nutritional value of indigenous fodder tree leaves i.e., chemical composition, macro mineral profile, secondary metabolites and digestion kinetics.

Materials and Methods

Harvesting and preparation of tree leaves: Leaves of *Morus alba*, *Acacia nilotica*, *Syzygium cumuni* and *Ziziphus jujuba* (Table 1) were collected from district Faisalabad of Punjab province, Pakistan in April, 2008. Sixteen indigenous fodder trees, four for each species, were harvested. Each tree was sampled from five sites (east, west, north, south and canopy). The collected leaves were dried separately in forced air oven at 55^{o}C, ground to pass a 2 mm sieve in Willey mill and saved in polythene bags for further analysis.

Chemical analysis: Dry matter (DM) content was determined by drying the sample at 105^{o}C in forced air oven till the constant weight. Ash content was measured after igniting sample in a muffle furnace at 550^{o}C for 4 hours (h). Crude protein (CP) was determined by Kjeldahl method (Anon., 1995). Neutral detergent fiber (NDF), acid detergent fiber (ADF) and acid detergent lignin (ADL) were determined by methods of Van Soest *et al*., (1991) without the use of alpha amylase but with use of sodium sulfite. Following the wet digestion (nitric acid and perchloric acid), the P was determined by the spectrophotometer (U 1100, Hitachi), while the flame photometer (Jenway PFP7) was used to estimate Na and K concentration. The Ca and Mg concentrations were determined colorimetrically. All chemical analyses were done in triplicate. Digestible energy (DE) was calculated by determining the gross energy (Harris, 1970) of tree leaves and residues of leaves at 48 h of incubation of *in sacco* trial. This estimation of DE was further used for the calculation of ME contents by following equation (Anon., 2001). ME = 1.01 x DE–0.45.

Total phenolics were determined by using Folin-Ciocalteu reagent method as described by Ainsworth & Gillespie (2007). All the samples were extracted in methanol. In 100mL of each sample 200uL of F-C reagent was added and vortex thoroughly 800uL of 700 mM Na_2CO_3 into each sample and incubated at room temperature for 2 h. 200uL sample was transferred to a clear 96-well plate and absorbance of each well was measured at 765nm. Amount of total phenolics was calculated using a calibration curve (R^2: 99.27) for gallic acid. The results were expressed as gallic acid equivalent per dry matter (Ainsworth & Gillespie, 2007). Tannins were determined by vanillin hydrochloride method. One gram of the ground plant material was extracted in 50 mL of methanol, mix occasionally and after 24 h centrifuge at 12000x*g* and supernatant was collected. In one mL of supernatant added 5 mL vilillin hydrochloride reagent and mix incubated at room temperature and noted the absorbance at 500 nm on microquant

spectrophotometer (BioTech, USA). The calibration curve catechin was used as standard for the calculation of tannins (Thimmaiah, 2004). Saponin was estimated by the method reported by Saddiqui & Ali (1997). For the determination of alkaloids, 5 g of powdered plant material was accurately weighed. It was made into a paste with 5% Sodium carbonate solution. Transferred into a flask and added 75 mL of chloroform in it, and refluxed for 15 minutes, then it was cooled and filtered. Filtrate was transferred to a separator, 40 mL of 5% Sodium carbonate solution was added, and agitated gently for 7 minutes. Chloroform layer was taken off and reduced to the volume of about 10 mL by distillation. 40ml of 1% suphuric acid was added, and extracted with two 20 mL volume of chloroform. Separating funnel separated aqueous phase. It was made alkaline with Ammonium hydroxide, and extracted with 10 mL portions of chloroform. Chloroform layers were combined, and washed with 5 mL of water. The total volume of fraction obtained was reduced to 5 mL by distillation. Chloroform was transferred to a crystallizing dish and remove remainder of chloroform through evaporation in a vacuum hood. Two mL of absolute alcohol was added to residue and evaporated to dryness at 100˚C and solid residue obtained was crude alkaloid.

$$\frac{\text{Weight of alkaloids obtained}}{\text{Total weight of sample}}$$

***In situ* trial:** An *in sacco* trial was conducted to examine the digestion kinetics of leaves of selected tree species. Nylon bags measuring 10×23 cm with an average pore size of 50 µm were used to determine digestibility, rate of disappearance, lag time and extent of digestion of DM and NDF. A mature ruminally cannulated buffalo bull (350±10kg) was used to study *In sacco* digestion kinetics of tree leaves. The bull was fed a blend of berseem fodder (*Trifolium alexandrinum*) and concentrate along with leaves of these trees to meet the nutritional requirements for 22 days. Initial 10 days were adjustment phase whereas following 12 days were for data collection. For each time point, there were 3 bags. Ground samples were weighed (5 g on DM basis) into nylon bags. The bags were manually pushed deep into the liquid phase of the ventral sac of rumen and incubated for 0, 3, 6, 12, 24, 36, 48 and 72 h. The bags were placed in the rumen in a reverse sequence. All bags were removed at the same time to reduce variation associated with the washing procedure. After removal from the rumen, the bags were washed in running tap water until the rinse was clear. These bags were dried in forced air oven at 60^{o}C for 48 h. The bags were weighed and the residues were transferred into bottles and stored for analysis. Digestibility was calculated at 48 h of incubation. Rate of disappearance was determined by subtracting the indigestible residue i.e. the 72 h residue from the amount in the bag at each point and then regressing the natural log (ln) of that value against time. Extent of digestion was determined at 72 h of incubation. Lag time was determined by the equation:

$$\text{Lag time (h)} = (\ln 100) - \text{Intercept} / \text{Rat of digestion}$$

Statistical analysis: The data collected regarding digestibility, lag time, rate of digestion and extent of digestibility were analyzed for variance analysis in completely randomized design and means were compared using computer software Anon., (1999).

Results and Discussion

Chemical composition: Chemical composition of tree leaves is presented in Table 1. The DM contents varied from 25% in *M. alba* to 47% in *S. cumuni*. The OM contents were 90.0, 92.0, 94.0 and 91.0% in *M. alba, A. nilotica, S. cumuni* and *Z. jujuba* tree leaves, respectively. *M. alba* contained highest CP (23%). Kandylis *et al*., (2009) reported higher levels of CP in *M. alba*. It is attributed that *M. alba* leaves have an appreciable potential as protein source in ruminant feeding. *M. alba* showed the maximum CP, therefore, may be used as supplementary protein source for ruminants (Yao *et al*., 2000). Lowest CP was observed in *S. cumuni* (8%). Rumen fermentation is affected if the CP level in diet is less than 10% (Alam & Djajanigra, 1994), however, CP level in these trees is higher than this level except *S. cumuni*. Differences in CP contents between leaves of different trees are probably due to differences in protein accumulation in them during growth. The NDF contents of *Z. jujuba* were highest (32%) and lowest in *A. nilotica* (25%). Schmidek *et al*., (2000) reported that NDF contents were from 30.2 to 39.3%, ADF from 17.2 to 21.7% and hemicelluloses from 11.0 to 22.0% of DM in three clones of *M. alba* leaves. The ADF contents were 16, 16, 23 and 17% in *M. alba, A. nilotica, S. cumuni* and *Z. jujuba*, respectively. Lower values of ADF in these tree leaves indicate good potential a ruminant feed (Bakshi & Wadhwa, 2007). *M. alba, A. nilotica, S. cumuni* and *Z. jujuba* contained 7, 4, 7 and 4% ADL, respectively. Shayo (1997) reported that *M. alba* leaves contained 8.1% lignin. Highest hemicellulose (15%) was observed in *Z. jujuba* and lowest in *S. cumuni* (5%). The hemicellulose content of most of the tree leaves vary between 10 to 15%. *M. alba, A. nilotica, S. cumuni* and *Z. jujuba* contained 10, 8, 6 and 9% ash, respectively, while ash contents of most of the tree leaves varied from 6 to 15% indicating rich source of minerals in previous studies (Mandal, 1997). The ME contents (available at 48 h of incubation) were 10.5, 10.2, 6.5 and 8.2 MJ/kg in *M. alba, A. nilotica, S. cumuni* and *Z. jujuba*, respectively. The ME contents more than 8.00 MJ/kg of DM except *S. cumuni*, indicates their potential use for ruminants.

The chemical composition of tree leaves vary with the maturity of leaves and also with localities (Mandal, 1997). Thus, the chemical composition of the leaves of plant species analyzed here provides a good source to be used as the nutrient source of ruminant animal feed.

Mineral composition: As given in Table 2, Ca concentration was higher and varied from 2.40% for *S. cumuni* to 3.80% for *A. nilotica*. Concentration of Ca in most of the tree leaves varied from 2 to 4%. Concentration of P ranged from 0.09% for *Z. jujuba* to 0.57% for *M. alba* and was lower than minimum animal requirements of P (0.2% of DM) reported by McDowell *et al*., (1984). Saha & Gupta (1987) reported that tree leaves are rich in Ca and poor in P. The Ca:P ratio in present study was wide (6.32, 8.44, 24.0 and 40.0 in *M. alba, A. nilotica, S. cumuni* and *Z. jujuba*, respectively) as compared to those recommended for ruminants (McDowell, 1997) and it should be adjusted to normal one (2:1 to 4:1) by supplementation with P for their proper utilization in the animal system (McDowell, 1992) when tree leaves are considerable part of ruminant ration. Ruminant can tolerate Ca:P ratio as wide as 7:1 (Anon., 1985) and higher Ca:P ratio reduces absorption of P (Anon., 2001). The Mg value varied from 0.72% in *M. alba* to 0.96% in *Z. jujuba* and can fulfill requirement (0.12-0.18% of diet DM) of ruminants (Anon., 1985). The Na concentration ranged from 0.15% in *A. nilotica* to 0.54% in *Z. jujuba*. Concentration of K was highest in *M. alba* (1.75%) and lowest in *Z. jujuba* (1%). Higher

mineral contents of the plants species in this study further provoke their use a good alternative for animal feed.

Secondary metabolites: Total phenolics concentration ranged from 0.93% for *M. alba* to 3.80% for *A. nilotica* (Table 4). Concentration of tannins varied from 1.27% for *M. alba* to 3.61% for *A. nilotica*. Alkaloids concentration was 0.19 and 0.32% for *Z. Jujuba and A. nilotica*, respectively. Saponin was observed only in *A. nilotica* (0.58%). Intake of plant secondary metabolites at high level reduces the nutrient utilization, feed efficiency and animal productivity, however, in the present study observed level of secondary metabolites was within safe range.

Tannins bind to proteins in the mouth reducing the palatability of the feed and subsequently decrease feed intake. Tannins have ability to bind and inhibit the digestive enzyme activities (Kumar & Singh, 1984) and affect the microbial and enzyme activities (Makkar *et al*., 1989), whereas, lower concentration of tannins can improve nutrition for ruminants by reducing protein degradation in the rumen and increasing the flow of amino acids to the intestine (Mc Nabb *et al*., 1996). Moreover, tannins have adverse impact on intestinal nematodes and nematode larvae (Waghorn, 1996). In this study, tannin level was below 4% of DM of fodder tree leaves. Concentration of tannins less than 4% in the ration is beneficial by promoting bypass protein and bloat suppression in ruminant animals (Aganga & Tshwenyane, 2003). Tannin concentrations higher than 5% adversely affect forage intake and digestibility (Perevolotsky *et al.*, 1993; Silanikove *et al*., 1996).

***In situ* dry matter digestion kinetics:** The DM digestibility at 48 h of incubation was highest (90.2%) for *M. alba* and lowest (54.8%) for *S. cumuni* (Table 3). Tree leaves were found to have *in situ* DM digestibility values above 54% indicating their potential for use in ruminant ration. Bakshi & Wadhwa (2007) reported higher *in vitro* digestibility (80.3%) of *M. alba*. High level of CP results in increased ruminal ammonia N concentration (Hristov *et al.*, 2004). Increased ruminal ammonia N status enhances microbial activity and growth, resulting in greater DM digestibility (Griswold *et al*., 2003). Highest DM digestibility observed with *M. alba* appears to be due to its high level of CP and lower level of NDF (26%), as asserted by Wiedmeier *et al*., (1983) for wheat straw. The DM lag time was shorter (0.63 h) for *A. nilotica* and greater (0.82 h) for *S. cumuni*. Rate of DM disappearance was higher (6.38% per h) for *A. nilotica* and lower (5.34% per h) for *S. cumuni*. Higher rate of DM disappearance for *A. nilotica* might be due to its shorter (0.63 h) lag time. Sarwar *et al*., (1996) reported that rate of DM disappearance highest in legume and the lowest in wheat straw. Extent of DM digestion at 72 h of incubation was maximum (98.26%) for *M. alba* and minimum (60.40%) for *S. cumuni*.

***In situ* neutral detergent fiber digestion kinetics:** The NDF digestibility at 48 h of incubation was highest (84.10%) for *M. alba* while lowest (17.58%) for *S. cumuni* (Table 4). Low NDF digestibility of *S. cumuni* might be due to its low CP and high lignin contents. The NDF lag time was shorter (0.71 h) for *A. nilotica* and greater for (0.98 h) for *S. cumuni*. Greater lag time may be due to nature of cell wall. Rate of NDF disappearance was highest (6.24% per h) for *A. nilotica* and lowest (4.92% per hr) for *S. cumuni*. The differences in rate of different fodder tree leaves may be due to chemical or physical nature of the fiber. Extent of NDF digestion at 72 h of incubation was maximum (96.8%) for *M. alba* and minimum (24.8%) for *S. cumuni*. Lower extent of *S. cumuni* NDF digestion might be due to its higher level of ADL and lower N contents.

Table 2. Chemical composition of fodder tree leaves.

Items	*M. alba*	*A. nilotica*	*S. cumuni*	*Z. jujuba*
Dry matter (%)	25.0	35.1	47.0	45.1
Organic matter (%)	90.0	92.0	94.0	91.0
Crude protein (%)	23.0	13.2	8.0	15.1
Neutral detergent fiber (%)	26.2	25.0	28.0	32.0
Acid detergent fiber (%)	16.0	16.0	23.0	17.0
Hemicellulose (%)	10.2	9.0	5.0	15.0
Acid detergent lignin (%)	7.0	4.0	7.0	4.0
Ash (%)	10.0	8.0	6.0	9.0
Metabolizable energy (MJ/kg)	10.5	10.2	6.0	8.2

Table 3. Mineral composition of fodder tree leaves (%).

Items	*M. alba*	*A. nilotica*	*S. cumuni*	*Z. jujuba*
Calcium	3.60 ± 0.11	3.80 ± 0.07	2.40 ± 0.09	3.60 ± 0.12
Phosphorus	0.57 ± 0.03	0.45 ± 0.03	0.10 ± 0.002	0.09 ± 0.001
Magnesium	0.72 ± 0.04	0.84 ± 0.05	0.72 ± 0.05	0.94 ± 0.04
Sodium	0.20 ± 0.01	0.15 ± 0.004	0.51 ± 0.03	0.54 ± 0.03
Potassium	1.75 ± 0.04	1.10 ± 0.07	1.33 ± 0.06	1.0 ± 0.02

Table 4. Secondary metabolites in tree leaves (%).

Items	*M. alba*	*A. nilotica*	*S. cumuni*	*Z. jujuba*
Total phenolics	0.93 ± 0.06	3.80 ± 0.11	2.85 ± 0.08	2.46 ± 0.1
Tannins	1.27 ± 0.04	3.61 ± 0.12	2.03 ± 0.05	2.75 ± 0.09
Alkaloids	Nil	0.32 ± 0.03	Nil	0.19 ± 0.02
Saponin	Nil	0.58 ± 0.05	Nil	Nil

Table 5. *In situ* dry matter digestion kinetics of fodder tree leaves.

Parameters	*M. alba*	*A. nilotica*	*S. cumuni*	*Z. jujuba*	SE
Digestibility[1]	90.2^{a}	65.6^{b}	54.8^{c}	65.2^{b}	3.92
Lag time (h)	0.76^{b}	0.63^{d}	0.82^{a}	0.71^{c}	0.02
Rate[2]	6.43^{a}	6.38^{a}	5.34^{c}	5.82^{b}	0.12
Extent[3]	98.26^{a}	66.40^{b}	60.40^{c}	67.90^{b}	4.45

Means within row with same superscripts are not statistically different ($p<0.05$)
[1]Dry matter digestibility (%) was determined at 48 h of incubation
[2]Rate of disappearance, %/h
[3]Extent of digestion (%) was determined at 72 h of incubation
SE stands for standard error

Table 6. *In situ* neutral detergent fiber digestion kinetics of fodder tree leaves.

Parameters	*M. alba*	*A. nilotica*	*S. cumuni*	*Z. jujuba*	SE
Digestibility[1]	$84.10^{\mathbf{a}}$	$53.11^{\mathbf{b}}$	$17.58^{\mathbf{c}}$	$53.34^{\mathbf{b}}$	7.17
Lag time (h)	$0.82^{\mathbf{b}}$	$0.71^{\mathbf{c}}$	$0.98^{\mathbf{a}}$	$0.76^{\mathbf{bc}}$	0.04
Rate[2]	$5.62^{\mathbf{b}}$	$6.24^{\mathbf{a}}$	$4.92^{\mathbf{c}}$	$5.99^{\mathbf{a}}$	0.25
Extent[3]	$96.8^{\mathbf{a}}$	$57.7^{\mathbf{c}}$	$24.8^{\mathbf{d}}$	$68.0^{\mathbf{b}}$	7.76

Means within row with same superscripts are not statistically different ($p<0.05$)
[1]Dry matter digestibility (%) was determined at 48 h of incubation
[2]Rate of disappearance, % per h
[3]Extent of digestion (%) was determined at 72 h of incubation
SE stands for standard error

Conclusion

Selected tree leaves by virtue of having high CP, shorter lag time and faster digestion rate are compatible to concentrates. A wider Ca to P ratio suggests supplementing cereal byproducts having high level of P along with these tree leaves when fed to ruminants. Based upon the higher availability of ME, CP and digestion kinetics, species trees for use as supplement in ruminant animals feed are ranked as *M. alba>A. nilotica=Z. jujuba>S. cumuni.*

References

Aganga, A.A. and S.O. Tshwenyane. 2003. Feeding value and anti-nutritive factors of forage tree legumes. *Pak. J. Nutr*., 2: 170-177.

Ainsworth, E.A. and K.M. Gillespie. 2007. Estimation of total phenolic content and other oxidation substrates in plant tissues using Folin-ciocalteu reagent. *Nature Protocols*, 2: 875-877.

Alam, M.P. and A. Djajanigara. 1994. Nutritive value and yield of potential tree leaves and shrubs in Bangladesh. In: *Proceedings of 7th AAAP Animal Science Congress on Sustainable Animal Production and Environment.* (Ed.): A. Sukmawati. Vol.2, held at Bali, Indonesia from July 6 to 11, 1994. 317-318.

Anonymous. 1985. National Research Council, *Nutrient requirements of sheep:* Sixth Revised Edition. National Academy Press, Washington, DC.

Anonymous. 1995. *Official Method of Analysis*, 16th ed. Animal Feeds: Association of Official Analytical Chemists, Arlington, VA, USA, pp. 1–18.

Anonymous. 1999. SPSS Uses guide. Release 10.0.1 edition. SPSS Inc.

Anonymous. 2001. National Research Council. *Nutrient requirements of dairy cattle*. Seventh revised edition. National Academy Press, Washington DC.

Anonymous. 2006. Smallstocks in development. *NR International Managers of the Livestock Production Programme (LPP),* http://www.smallstock.info/info/feed/forage.htm (accessed 28 November, 2005)

Bakshi, M.P.S. and M. Wadhwa. 2007. Tree leaves as complete feed for goat kids. *Small Rumin. Res*., 69: 74-78.

Baumer, M. 1992. Trees as browse and to support animal production. In: *Legume trees and other fodder trees as protein sources for livestock.* (Eds.): A. Speedy, P.L. Pugliese. FAO Animal Production and Health Paper No. 102. FAO (Food and Agriculture Organization), Rome, Italy, pp. 1-10.

Bonsi, M.L.K., P.O. Osuji and A.K. Thuah. 1995. Effect of supplementing tef straw with different level of *Leucaena* or *Sasbania* on the degradability of tef straw, sesbania, leucaena, tagaste and vernonia and certain rumen and blood metabolities in Ethopian menz sheep. *Anim. Feed Sci. Technol*., 52: 101-129.

Griswold, K.E., G.A. Apgar, J. Bouton and J.L. Firkins. 2003. Effects of urea infusion and ruminal degradable protein concentration on microbial growth, digestibility, and fermentation in continuous culture. *J. Anim. Sci*., 81: 329-36.

Harris, L.E. 1970. *Nutrition Research Techniques for Domestic and Wild Animals*. Vol. 1. Animal Science Department, Utah State University, Logan, USA.

Hristov, A.N., R.P. Etter, J.K. Ropp and K.L. Grandeen. 2004. Effect of dietary crude protein level and degradability on ruminal fermentation and nitrogen utilization in lactating dairy cows. *J. Anim. Sci*., 82: 3219-3229.

Kandylis, K., I. Hadjigeorgiou and P. Harizanis. 2009. The nutritive value of mulberry leaves (Morus alba). *Trop. Anim. Health Prod*., 41: 17-24.

Kumar, R. and M. Singh. 1984. Tannins: their adverse role in ruminant nutrition. *J. Agric. Food Chem*., 32: 447-453.

Makkar, H.P.S., B. Singh and S.S. Negi. 1989. Relationship of rumen degradability with microbial colonization, cell wall constituents and tannin levels in some tree leaves. *Anim. Prod*., 49: 299-303.

Malik, M.Y., A.A. Sheikh and W.H. Shah. 1967. Chemical composition of indigenous fodder tree leave. *Pak. J. Sci.*, 19: 5-7.

Mandal, L. 1997. Nutritive values of tree leaves of some tropical species for goats. *Small Rumin. Res*., 24: 95-105.

Mc Nabb, W.C., G.C. Waghorn, J.S. Peters and T.N. Barry. 1996. The effect of condensed tannins in *Lotus pedunculatus* on the stabilization and degradation of ribulose-1,5-bisphosphate carboxylase (EC 4.1.1.39: rubisco) protein in the rumen and the sites of rubiso digestion. *Br. J. Nutr*., 76: 535-549.

McDowell, L.R. 1992. *Minerals in Animal and Human Nutrition*. San Diego, CA: Academic Press, pp. 49-51.

McDowell, L.R. 1997. *Minerals for Grazing Ruminants in Tropical Regions*. University of Florida. Gainesville.

Niderkorn, V. and R. Baumonta. 2009. Associative effects between forages on feed intake and digestion in ruminants. *Animal*, 3: 951-960.

Perevolotsky, A., A. Brosh, O. Ehrilich, M. Gutman, Z. Henkin, and Z. Holtezer. 1993. Nutritional values of common oak (*Quercus calliprinos*) browse as fodder for goats: Experimental results in ecological perspective. *Small Rumin. Res*., 11: 95-106.

Raghuvansi, S.K.S., R. Prasad, A.S. Mishra, O.H. Chaturvedi, M.K. Tripathi, A.K. Misra, B.L. Saraswat and R.C. Jakhmola. 2007. Effect of inclusion of tree leaves in feed on nutrient utilization and rumen fermentation in sheep. *Biores. Technol*., 98: 511-517.

Reddy, D.V. 2006. *Production of forage in the Indian context*. In: *Fodder Production and Grassland Management for Veterinarians*. Oxford & IBH Publishing Co. Pvt. Ltd. New Delhi, pp 453.

Saha, S.C. and B.N. Gupta. 1987. Tree leaves as feed for dairy cattle in India. *India Dairyman*, 39: 489-492.

Schmidek, A., R. Takahashi, A.N. de Medeiros and K.T. de Resende. 2000. Bromatological composition and degradation rate of mulberry in goats. In: *FAO Electronic Conference on Mulberry for Animal Production* (Morus1-L); http://www.fao.org/ag/AGA/AGAP/ FRG/Mulberry/Posters/PDF/Schmidk1.pdf (accessed 29 November 2009).

Shayo, C.M. 1997. Uses, yield and nutritive value of mulberry (*Morus alba*) trees for ruminants in the semi-arid areas of central Tanzania. *Trop. Grasslands*, 31: 599-604.

Siddiqui, A.A. and Ali. 1997. *Practical Pharmaceutical Chemistry*. 1st Edition. CSB Publisher and Distributors, 4596/1A, 11-Dariganj, New Dehli, India: 125-131.

Silanikove, N., N. Gilboa, I. Nir, A. Perevolotsky and Z. Nitsan. 1996. Effect of a daily supplementation of polyethylene glycol on intake and digestion of tannin-containing leaves (*Quercus calliprinos*, *Pisticia lentiscus* and *Ceratonia siliqua*) by goats. *J. Agric. Food Chem*. 44: 199-205.

Singh, R.V. 1982. *Fodder Trees of India*. Oxford and IBH Publishing, New Delhi, pp. 122: 139, 199.

Soliva, C.R., M. Kreuzer, N. Foidl, G. Foidl, A. Machmüller and H. D. Hess. 2005. Feeding value of whole and extracted *Moringa oleifera* leaves for ruminants and their effects on ruminal fermentation *In vitro*. *Anim. Feed Sci. Technol*., 118: 47-62.

Thimmaiah, S.R**.** 2004. *Standard methods of Biochemical Analysis*. New Dehli, India: Kalyani Publishers.

Van Soest, P.J., B. Rebortson and B.A. Lewis. 1991. Method for dietary fiber, neutral detergent fiber and non starch poly saccharides in relation to animal nutrition. *J. Dairy Sci*., 74: 3583-3597.

Waghorn, G.C. 1996. Condensed tannins and nutritive absorption from small intestine. Page 175 in *Proc. Can. Soc. Anim. Sci*., Lethbridge, Alberta, Canada. L. M. Rode, ed.

Wiedmeier, R.D., J.R. Males and C.T. Gaskin. 1983. Effect of dietary crude protein on the dry matter digestibility of wheat straw in cattle. *J. Anim. Sci*., 57: 1568-1575.

Yao, J., B. Yan, X.Q. Wang and J.X. Liu. 2000. Nutritional evaluation of mulberry leaves tas feeds for ruminants. *Livest. Res. Rural Develop*., 12: 9-16.

(Received for publication 5 January 2010)