



Effect of Feeding *Artocarpus heterophyllus*, *Terminalia bellerica* and *Carica papaya* Plant Leaves on Rumen Microbial Enzymes and Growth Performances in Assam Hill Goat

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ABSTRACT

In this study, three medicinal plants viz. *Artocarpus heterophyllus* (Jackfruit), *Terminalia bellerica* (Bhumura) and *Carica papaya* (Papaya) were investigated for their effect on the rumen enzyme profiles and growth performances in Assam hill goat. A total of eighteen (18) Assam Hill (local) goats of 3-4 months of age were procured and reared in the Experimental Animal Shed, Department of Veterinary Physiology, College of Veterinary Science, Assam Agricultural University, Khanapara, Guwahati, Assam, India. The animals were divided into three groups viz. control receiving substrate ration, T₁ with plant leaves @ 5 % level and T₂ groups @ 10 % level of total mixed ration (TMR). The duration of the experiment was 90 days (from March, 2016 to May, 2016). The fibre degrading microbial enzymes viz. carboxymethyl cellulase, endoxylanase, β- glucosidase were estimated in rumen liquor of the animals during the study period and rumen liquor analysis revealed that out of the three rumen fibrolytic enzymes, Carboxymethyl cellulase showed better (P<0.05) enzyme activity in T₂ group (2.83 ± 0.01 μmol glucose/ml/h). The average body weight (kg per animal) recorded in the present experiment showed significant trend (P<0.01) from 0 to 90 days of experiment where higher value (9.62 ± 0.43 kg/animal) was obtained in T₂ group. From this study, it can be concluded that these three medicinal plants viz., *A. heterophyllus*, *C. papaya*, *T. bellerica* do not have any harmful effect on normal functioning of the rumen. Thus, the plant leaves can be incorporated in the diet up to 10% of the total mixed ration.

HIGHLIGHTS

- The medicinal plants viz., *A. heterophyllus*, *T. bellerica* and *C. papaya*, are important for normal functioning of the rumen of animals.
- These leaves can be incorporated in the diet of animals up to 10 % of the total mixed ration.

Keywords: Assam hill goat, enzyme, plant, rumen, total mixed ration

Livestock production is a vital component of the Agricultural Industry in India. About 20.5 million people in India depend upon livestock for their livelihood and much effort is being directed towards improving this sector. Goats (*Capra hircus*) are the dominant small ruminants that play a significant role in the rural economy

of India. India possesses second largest goat population in

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the world which is 135.2 million. It provides an alternative source of livelihood to the farmers all the year round. In Assam, the population of goats is 61.69 lakhs (19th Livestock Census, 2012) and mostly reared by the poor and landless farmers of rural areas. The main constrain to goat production is the lack of feed, particularly in the dry season that affects the productivity (Phengsavanh, 2003; Xayph, 2005). Commercial concentrates are generally used as supplements to goat. Considering the cost and availability of concentrates, it is wise to use it most judiciously. As an alternate source, tree leaves, green forages are used to overcome the feed gap that arises from seasonal fluctuation. Jackfruit (*Artocarpus heterophyllus*), Bhumura (*Terminalia bellerica*) and Papaya (*Carica papaya*), and are the locally available trees in Assam and are also relished by goat. Leaves of these trees can be used as a green fodder during drought period. Studies have shown that these medicinal plant leaves possess antihelmintic, antiprotozoan, antibacterial, antifungal, antiviral, anti-inflammatory, antihypertensive, hypoglycemic and hypolipidemic, wound healing, antitumor, free-radical scavenging, antisickling, neuroprotective, diuretic, abortifacient and antifertility activities (Yadav and Agarwala, 2011; Maisarah *et al.*, 2013; Raaman, 2015) as they are rich source of phytochemicals such as tannin, saponin, phenolic compounds, alkaloids, flavonoid, carotenoid, cardiac glycosides, steroid, ascorbic acid, vitamin E, papain enzyme etc. (Varisha *et al.*, 2013). But the information related to the potential of these tree leaves along with substrate feed in respect of rumen microbial enzyme activities including carboxymethyl cellulase, β -glucosidase and xylanase and their effect on growth performance of goat is very limited. Therefore, the present experiment was conducted in order to understand about the changes of rumen microbial enzymes and growth performances in Assam hill goat by feeding locally available plants along with substrate ration.

MATERIALS AND METHODS

The experiment was conducted in the Department of Veterinary Biochemistry, College of Veterinary Science, Assam Agricultural University, Khanapara, Guwahati-781022 under the DBT Twinning Project (NER) entitled “Role of plants as a modifier of rumen to reduce methane production and improve productivity in small animals” during the study period (from March, 2016 to May, 2016).

Collection of plant materials

For the present study, fresh and disease free leaves of three locally available medicinal plants *Artocarpus heterophyllus* (Jackfruit), *Terminalia bellerica* (Bhumura) and *Carica papaya* (Papaya) were collected from six different places in and around Guwahati, Assam, India. These leaves were collected based on palatability and ease of availability.

Processing of plant leaves

The plant materials were washed under running tap water followed by distilled water to remove dust and then cut into small pieces, dried under sun and pulverized into fine powder in a grinding machine. The powder was then kept in small plastic bags away from the light, heat, moisture with proper labelling till further analysed.

Experimental animals

The experimental animals were reared in Experimental Animal Shed, Department of Veterinary Physiology, College of Veterinary Science, Assam Agricultural University, Khanapara, Guwahati-781022, Assam, India. The experiment was conducted for a period of 90 days (from March, 2016 to May, 2016). A total of eighteen Assam hill (local) kids of 3-4 months of age and average body weight of 6-8 kg were procured locally. Prior to the arrival of the procured goats, the pens and surrounding environment were thoroughly cleaned and disinfected with anti-septic solutions. The animals were conditioned for a period of 15 days prior to start of the actual experiment and given standard ration/ substrate feed, green grasses. During the study period, the animals were given prophylactic treatment consisting of deworming for endo and ecto parasites. Everyday animal sheds, wooden floors, watering and feeding pans were cleaned to avoid microbial contaminations. The experimental animals were housed in a well-ventilated goat shed having a wooden floor. Goats were maintained under semi-intensive system. Animals were given substrate feed two times in a day (morning and evening) and water was given *ad libitum*. After feeding they were turned out for natural grazing from 9.00 A.M. to 12.30 P.M. and again from 2.30 P.M. to 5.30 P.M. This schedule was followed till the end of the experiment.

Design of the experiment

The animals were divided into three (3) groups *viz.* Control (Substrate feed), T₁ (Plant leaves @ 5 % level of Total Mixed Ration) and T₂ (Plant leaves @ 10 % level of Total Mixed Ration) containing six (6) animals in each group irrespective of sex.

Experimental dietary treatment

Substrate ration or feed was prepared using conventional feed ingredients *viz.* maize, wheat bran, ground nut cake, mineral mixture, salt and vitamins (Table 1). The calculated nutritive value of the substrate feed was 13.89 % DCP and 78.89 % TDN. The three medicinal plant leaves *viz.* *Artocarpus heterophyllus* (Jackfruit), *Terminalia bellerica* (Bhumura) and *Carica papaya* (Papaya) were mixed in the ratio of 1:1:1 and mixed in the substrate feed at concentration 5 % and 10 % level.

Table 1: Composition of the ration given to the animals with or without medicinal plant leaves

Ingredients	Control	T ₁ (5 % level)	T ₂ (10 % level)
Maize	65	56	60
Wheat bran	20	25	20
GNC	12	11	7
Mineral mixture	2	2	2
Salt	1	1	1
Medicinal leaves	—	5	10
Total	100	100	100

Collection and processing of rumen liquor

Rumen liquor was collected as per the standard method (Singh and Kundu, 2010). After collection, rumen liquor was processed as per the standard method (Singh *et al.*, 2011) for estimation of fibre degrading enzymes *viz.* carboxymethyl cellulase, β -glucosidase and xylanase.

Estimation of carboxymethyl cellulase enzyme activity

Carboxymethyl cellulase is also known as endoglucanase. The method described by Miller (1959) was used to estimate the carboxymethyl cellulase enzyme activity.

Estimation of β -glucosidase (β -d-glucoside glucohydrolase) enzyme activity

The enzyme catalyses the hydrolysis of cellobiose and short chain oligosaccharides to release glucose. The activity was measured as per the method described by Shewale and Sadana (1978).

Estimation of xylanase (1, 4- β -xylan xylano hydrolase) enzyme activity

The enzyme catalyses the hydrolysis of xylan and releases D-xylose. The activity was measured as per the method described by Miller, 1959.

Weighing of the animals

The body weight of the individual animal was recorded at 7.00 A.M. (before offering feed and water) from beginning of the experiment (0 day) till the end of the experiment (90 days) at 30 days interval.

Statistical analysis

The statistical analysis of the experimental data were carried out according to the method described by Snedecor and Cochran (1994).

RESULTS AND DISCUSSION

The carboxymethyl cellulase enzyme activity (μmol glucose/ml/h) of all the three groups of Assam hill goat are presented in Table 2. The carboxymethyl cellulase enzyme activities (μmol glucose/ml/h) in rumen liquor of control, T₁ and T₂ groups of Assam local goat were recorded as 2.80 ± 0.01 , 2.77 ± 0.01 and 2.83 ± 0.01 respectively with overall mean value of 2.80 ± 0.01 μmol glucose/ml/h. The values of carboxymethyl cellulase enzyme recorded in the present experiment were in close agreement with the earlier reports (Agarwal *et al.*, 2002; Wang *et al.*, 2016). Analysis of variance revealed significant ($P < 0.05$) differences between T₁ and T₂ groups but no significant differences were observed between control and T₁ groups & also between control and T₂ groups of Assam local goat which may be attributed to the difference in fibre content of the ration supplied to T₁ and T₂ groups of Assam local goats at 5% and 10% level of total mixed ration respectively. A higher proportion of fibre content in the 10% level of

Table 2: Carboxymethyl cellulase, β - glucosidase and xylanase enzyme activity (Mean \pm SE) in rumen liquor of Assam hill goat

Groups	Carboxymethyl cellulase ($\mu\text{mol glucose/ml/h}$)	β – Glucosidase ($\mu\text{mol p-nitrophenol/ml/min}$)	Xylanase ($\mu\text{mol xylose/ml/min}$)
Control	2.80 ^{ac} \pm 0.01	0.35 ^{NS} \pm 0.01	2.90 ^{NS} \pm 0.01
T ₁	2.77 ^{ab} \pm 0.01	0.36 ^{NS} \pm 0.01	2.92 ^{NS} \pm 0.01
T ₂	2.83 ^c \pm 0.01	0.37 ^{NS} \pm 0.01	2.93 ^{NS} \pm 0.01
Overall Mean \pm SE	2.80 \pm 0.01	0.36 \pm 0.00	2.92 \pm 0.01

^{a, b, c} Means with different subscripts in a column differ significantly (P<0.05); ^{NS} Non-significant.

Table 3: Average body weight (Mean \pm SE) in different groups of Assam hill goats on different days of feeding

Groups	Average body weight per animal (kg)				Overall Mean \pm SE
	0 day	30 days	60 days	90 days	
Control	6.80 ^a \pm 0.20	7.20 ^b _A \pm 0.23	8.03 ^c _A \pm 0.20	8.93 ^d _A \pm 0.21	7.74 _A \pm 0.20
T ₁	6.98 ^a \pm 0.27	7.78 ^b _{AB} \pm 0.36	8.65 ^c _{AB} \pm 0.40	9.45 ^d _{AB} \pm 0.42	8.22 _B \pm 0.26
T ₂	7.20 ^a \pm 0.27	7.92 ^b _{BC} \pm 0.37	8.75 ^c _{BC} \pm 0.41	9.62 ^d _{BC} \pm 0.43	8.37 _{BC} \pm 0.26
Overall Mean \pm SE	6.99 ^a \pm 0.14	7.63 ^b \pm 0.19	8.48 ^c \pm 0.21	9.33 ^d \pm 0.21	

^{A, B, C} Means with different subscripts in a column differ significantly between groups (P<0.05); ^{a, b, c, d} Means with different superscript in a row differ significantly between days (P<0.01).

total mixed ration supplied to the T₂ group would have stimulated the microbial enzyme system responsible for its hydrolysis resulting in more ruminal enzyme activity of carboxymethyl cellulase (Vaithyanathan *et al.*, 2015). Lower carboxymethyl cellulase enzyme activity in T₁ group supplied with 5% level of total mixed ration contributed to their poor cellulose digestibility indicating significance of the rumen ciliated protozoa in fibre digestion (Santra and Karim, 2002; Kamra *et al.*, 2003; Agarwal *et al.*, 2004; Vaithyanathan *et al.*, 2015).

The β -glucosidase enzyme activity ($\mu\text{mol p-nitrophenol/ml/min}$) of all the groups of Assam hill goat are presented in Table 2. The β -glucosidase enzyme activities ($\mu\text{mol p-nitrophenol/ml/min}$) in rumen liquor of control, T₁ and T₂ groups of Assam local goat were recorded as 0.35 \pm 0.01, 0.36 \pm 0.01 and 0.37 \pm 0.01 respectively with overall mean value of 0.36 \pm 0.00 $\mu\text{mol p-nitrophenol/ml/min}$. Analysis of variance revealed non-significant differences amongst all the groups of Assam local goat in respect of β -glucosidase enzyme. These findings were in close agreement with the earlier reports (Agarwal *et al.*, 2002; Kamra *et al.*, 2003; Singh, 2004) where they have reported activity of β - glucosidase enzyme as 0.20 \pm 0.2 to 0.18 \pm 0.04 $\mu\text{mol pnp/min/ml}$. Therefore, the total mixed ration

supplied to T₁ and T₂ groups of Assam local goats did not alter the ruminal environment and digestion.

The xylanase enzyme activity ($\mu\text{mol xylose/ml/min}$) of all the groups of Assam hill goat are presented in Table 2. The xylanase enzyme activities ($\mu\text{mol xylose/ml/min}$) in rumen liquor of control, T₁ and T₂ groups of Assam local goats were recorded as 2.90 \pm 0.01, 2.92 \pm 0.01 and 2.93 \pm 0.01 respectively with overall mean value of 2.92 \pm 0.01 $\mu\text{mol xylose/ml/min}$ which were in close agreement with the earlier reports (Agarwal *et al.*, 2002; Wang and Wang, 2016). Statistically the xylanase enzyme activities ($\mu\text{mol xylose/ml/min}$) were found to be non-significant amongst control, T₁ and T₂ groups of Assam local goats. These findings were in close agreement with the earlier reports (Agarwal *et al.*, 2004; Vaithyanathan *et al.*, 2015; Wang and Wang, 2016) indicating no effect in the rumen micro flora and digestion in Assam local goats.

The average body weight (kg per animal) of control, T₁ and T₂ groups of Assam hill goats are presented in Table 3. The average body weight (kg per animal) recorded in the present experiment varied from 6.80 \pm 0.20 to 8.93 \pm 0.21 with overall mean value of 7.74 \pm 0.20 kg per animal for control, 6.98 \pm 0.27 to 9.45 \pm 0.42 kg per animal with overall mean value of 8.22 \pm 0.26 kg per animal for T₁,

7.20 ± 0.27 to 9.62 ± 0.43 kg per animal with overall mean value of 8.37 ± 0.26 kg per animal for T₂ groups of Assam hill goats respectively.

The average body weight (kg per animal) recorded in the present experiment showed increased significant trend (P<0.01) from 0 to 90 days of experiment in all the groups where higher value was obtained in T₂ group (9.62 ± 0.43 kg/animal) of Assam hill goat at 90 days of experiment which might be due to increase in age and also due to higher nutritive value of medicinal plant leaves at 10 % level of total mixed ration (Chukwuku *et al.*, 2013). The body weight values recorded in the present experiment showed significant differences (P<0.01) between 30, 60 and 90 days of experiment in all the groups and the values recorded during 30, 60 and 90 days of experiment were also found to be statistically significant (P<0.05) between control and T₂ groups of Assam hill goat but not between control and T₁ groups. From the experiment, it can be observed that T₂ group showed better result by feeding 10% level of leave mixture of the total mixed ration. These findings recorded in the present experiment were in close agreement with the earlier reports (Khaing *et al.*, 2015; Nath *et al.*, 2017).

CONCLUSION

From the present experiment, it could be concluded that out of the three fibre degrading enzymes *viz.* carboxymethyl cellulase, β-glucosidase and xylanase, the activity of carboxymethyl cellulase enzyme was found to be significantly (P<0.05) higher in T₂ group indicating higher fibre degradability in rumen and the growth performances in T₂ group were also found to be better (P<0.01) than control and T₁ groups of Assam hill goat. Finally it can be inferred that supplementing the total mixed ration for goat with 10% was better than 5% of the leave mixture.

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