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Dietary supplementation of *Ficus hookeri* leaves as herbal feed additive affects nutrient utilization and growth performances in growing crossbred calves

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ABSTRACT

The objective of this study was to evaluate the effect of dietary supplementation of Da qing shu (*Ficus hookeri*) leaf meal as herbal feed additive on nutrients utilization and growth performance of growing crossbred calves. Ten Jersey male cross-bred calves (~ 4 months of age) were divided equally into two groups (G1 and G2) and were fed *ad lib.* under stall feeding on a paddy straw based mixed ration consisting of 50% paddy straw and 50% concentrate mixture for a duration of 140 days. Two types (C1 and C2) of iso-nitrogenous concentrate mixtures were prepared. Four per cent (w/w) wheat bran in concentrate mixture (C2) of test group (G2) was replaced by sun dried ground *Ficus hookeri* leaf meal. Experimental calves of test group (G2) were fed *Ficus hookeri* leaf meal @ 2% of the diet. Daily dry matter intake (g/d) was similar among the calves of two experimental groups. Apparent digestibility of DM, OM, NDF, ADF and cellulose improved in the *Ficus hookeri* leaf meal fed calves (G2). DCP value of the ration was similar while, TDN value of the ration was higher in the calves of *Ficus hookeri* leaf meal supplemented group (G2). However, plane of nutrition among the calves of both experimental groups was similar. Average finishing body weight, daily body weight gain, feed conversion efficiency and blood glucose level were higher in *Ficus hookeri* leaf fed calves (G2). Therefore, it may be concluded from the results of the study that dietary supplementation of Da qing shu (*Ficus hookeri*) leaf meal as herbal feed additive @ 2% of total mixed diet significantly improved the fibre digestibility, average daily body weight gain and feed conversion efficiency in growing Jersey male crossbred calves.

Keywords: Calves, Feed additive, Feed conversion efficiency, *Ficus hookeri*, Growth, Nutrient utilization

Feed additives such as ionophores, defaunating agents, antibiotics have proved very effective in reducing dietary energy and protein losses; however, contemporary biosecurity threats have restricted their use in many European countries in animal feeds (Malik *et al*. 2019). Use of chemical feed additives in ruminant diets for improving nutrient utilization, is controversial now-a-days because of the risk of their transmission into meat and milk as well as the antimicrobial resistance, focussing the concern of new alternatives to chemical feed additives in the field of animal nutrition (e Silva *et al.* 2021, Swelum *et al.* 2021). In recent years, there has been an increased interest to use natural products containing plant secondary/ bioactive compounds instead of chemical feed additives to modify rumen fermentation for improving feed utilization and productive performances of ruminant animals (Taku *et al*. 2019, Bhatt *et al.* 2021, Uushona *et al*. 2022). Tropical plants normally contain wide spectrum of secondary compounds. Tree leaves contain different classes of bioactive compounds such as tannins, saponins, flavonoids and many other plant secondary metabolites which have been shown to potentially improve rumen microbial fermentation (Chaturvedi *et al.* 2021, Santra *et al*. 2021) and improve animal productivity (Santra *et al.* 2020, Jiao *et al.* 2021, Tilahun *et al.* 2022).

Several *in vitro* studies showed that plant secondary metabolites seem to have the potential to favourably manipulate rumen fermentation to reduce ruminal methane emissions and ciliated protozoal population for efficient use of dietary energy and protein (Bhatta *et al*. 2017, Singh *et al*. 2018, Malik *et al.* 2019, Akanmu *et al.* 2020). Lemon grass (*Cymbopogon citratus*), a medicinal herb contains essential oil citronella, which has proven beneficial effect on digestibility, rumen ecology, microbial population and protein synthesis and meat quality in ruminants (Bhatt *et al*. 2021). Phyto-chemicals present in the Australian Acacia leaves reduced the ruminal methanogenesis and improved the nutrient utilization, growth performances and meat quality in ruminants (Uushona *et al*. 2022). However, *in vivo* studies on plant secondary metabolites to use as natural feed additive to manipulate rumen fermentation for efficient utilization of dietary energy and protein to improve productivity of animals is almost lacking/very

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scanty particularly in India. Based on our previous *in vitro* studies in laboratory on methane and protozoa population reduction potential (Choudhary 2016), Da qing shu (*Ficus hookeri*) tree leaves not yet been investigated *in vivo* were selected for supplementation in the diet of calves as herbal feed additive. Therefore, the present experiment was conducted to study the effect of dietary supplementation Da qing shu tree leaf meal as herbal feed additive on nutrient utilization, plane of nutrition and growth performances of growing cross bred calves.

MATERIALS AND METHODS

Collection and processing of plant materials to use as herbal feed additive: Da qing shu (*Ficus hookeri*) tree leaves were collected from Barapani, Shillong, Tripura during the month of April. Both old and newly emerged type of leaves from trees were harvested and sun dried. The sun dried leaves were ground (2 mm size) in a hammer mill prior to the determination of chemical composition and subsequent use in concentrate mixture preparation.

Selection of experimental animals, dietary treatment and experimental design: Ten growing Jersey male cross-bred calves (age four months, body weight 91.8±1.37 kg) were randomly divided into two equal groups (G1 and G2) having five animals in each group on the basis of body weight so that average body weight of the two experimental groups was similar. The animals were dewormed at the beginning of the experiment. Two types (C1 and C2) of iso-nitrogenous concentrate mixtures were prepared (Table 1). Four per cent (w/w) wheat bran in concentrate mixture (C2) of test group (G2) was replaced with Da qing shu (*Ficus hookeri*) leaf meal (sun dried leaf powder). All the experimental calves were fed individually under stall feeding on a paddy straw based total mixed ration with roughage to concentrate ratio of 50:50 for a duration of 140 days to meet out maintenance and growth (600 g average daily gain) requirement (NRC 2001). Based on published literature (Malik *et al.* 2017, Yusuf *et al.* 2017, Jafari *et al*. 2018, Bhatt *et al.* 2021), calves of test group (G2) were fed *Ficus hookeri* leaf meal @ 2% of total diet as herbal feed additive. Concentrate mixture was offered once daily at 9:00 AM after discarding previous day's residue if any. Measured quantity of paddy straw was offered twice daily at 10.00 AM and 18.00 PM for an excess of 10% after discarding residue. Record of daily feed intake was maintained throughout the experimental period. All the experimental animals were weighed weekly to record their body weight changes during the experiment. Proper health management and sanitation conditions were maintained throughout the experimental period.

Evaluation of nutrient intake and its utilization: A digestion trial of ten days (i.e. 3 days adaptation followed by 7 days sample collection) was conducted on all the ten experimental crossbred calves after 120 days of experimental feeding. During the digestion trial, daily faeces voided, feed offered and residues left were recorded. Fresh samples of fodder/paddy straw and concentrate mixture offered, residues left, and faeces voided were collected daily in the morning after thorough mixing so as to obtain homogeneous and representative samples. 24 h faecal output of each animal was quantified and 25 % of the voided faecal sample was dried in forced draught hot air oven at 60°C. The dried faecal sample of individual animal for each day of the 7 days collection period was bulked, sampled and ground in a hammer mill to pass through 2 mm sieve. The ground samples were stored in air tight bottles until required for chemical analysis.

Blood samples: Blood samples were obtained from each

Table 1. Chemical composition (% DM) of experimental ration

Attributes	Concentrate mixture		Da qing shu (*Ficus hookeri*) leaf	Paddy straw
	C1	C2		
Physical composition (%)				
Da qing shu (*Ficus hookeri*) leaf	00	04	-	-
Maize grain	45	45	-	-
Wheat bran	29	25	-	-
Mustard cake	23	23	-	-
Mineral mixture	2	2	-	-
Common salt	1	1	-	-
Vitablend® AD_3	@ 20 g/100 kg concentrate mixture			
Chemical composition (% DM)				
Organic matter (OM)	93.7	93.4	84.6	86.6
Crude protein (CP)	18.2	17.7	10.2	3.4
Ether extract (EE)	4.7	4.5	3.7	1.9
Total carbohydrate (TCHO)	70.8	71.2	70.8	81.3
Neutral detergent fibre (NDF)	34.8	35.7	55.1	71.5
Acid detergent fibre (ADF)	11.8	12.1	37.1	41.1
Cellulose	7.6	7.8	26.4	34.3
Acid detergent lignin (ADL)	3.9	4.1	11.5	6.1
Acid insoluble ash			7.4	10.2

experimental cow at 0, 45 and 90 days of experimental feeding. Before offering feed, blood samples were collected from the jugular vein into 10 ml tube containing Na heparin as anticoagulant. Samples were centrifuged (5000 × g for 20 min at 4°C) with in 15 min, and collected plasma was frozen immediately at -20°C until analyzed.

Growth trial: The growth trial lasted for 140 days during which feed intake were recorded daily for each calf. Calves body weight were recorded for 2 consecutive days fortnightly immediately before offering feed and water and these values were used to determine body weight gain.

Chemical analysis: The feed offered, residues left and faeces voided were analyzed for DM by drying at 100°C for 24 h, OM by ashing at 550°C for 4 h and crude protein (CP) by Kjeldahl technique (AOAC 2005). NDF, ADF and ADL were estimated following the method of Van Soest *et al*. (1991). Cellulose content was calculated by subtracting ADL from ADF. Blood glucose (Cooper and Mc Daniel 1970), urea nitrogen (Rahamatullah and Boyde 1980), total protein and albumin (Annino 1976) were estimated in plasma of experimental animals.

Statistical analysis: Data were analyzed by means of SPSS software package version 20.0 following the normal statistical techniques. The groups mean were ranked with Duncan's multiple range test (Duncan 1955).

RESULTS AND DISCUSSION

*Chemical composition of experimental diet:*The chemical composition of concentrate mixture, Da qing shu (*Ficus hookeri*) leaf powder, and paddy straw indicated that the CP content in the concentrate mixture ranged from 17.7% to 18.2% while it was 10.2% and 3.4% in *Ficus hokkeri* leaf powder and paddy straw, respectively (Table 1). The overall nutrient composition of concentrate mixture (C1) for feeding to the control group (G1) and concentrate mixture (C2) for feeding to the test group (G2) was similar. The chemical composition of paddy straw used in the present experiment was comparable to the values reported earlier by Santra *et al.* (2013a). In the present experiment, Da qing shu (*Ficus hookeri*) leaf meal contained 10.2% crude protein which was lower than earlier reported value for *Ficus* sp. tree leaves (Khan *et al*. 2014).

Effect of feeding Ficus hookeri *leaf meal on feed intake, digestibility and nutrient utilization of crossbred calves:* All the experimental calves of control (G1) and test (G2) groups were maintained on a ration consisting of roughage/ paddy straw and concentrate mixture in the ratio of 50:50 under cafeteria system of feeding management. However, the actual intake of paddy straw and concentrate mixture ratio by the experimental calves in G1 and G2 experimental groups was 47.8:52.2 and 48.8:51.2, respectively. Daily dry matter intake in terms of per kg body weight as well as per kg metabolic body size was similar among the calves of two experimental groups (Table 2). The dry matter intake as % of body weight was 3.3 and 3.1% for the experimental calves of G1 and G2 groups, respectively

Table 2. Effect of dietary Da qing shu (*Ficus hookeri*) leaf supplementation as herbal feed additive on intake, nutrient digestibility and plane of nutrition in growing cross-bred calves

Attribute	Level of *Ficus hookeri* leaf meal (%)		SEM	P value
	0 (G1)	2 (G2)		
DMI				
Roughage (kg/d)	2.2	2.2	0.03	0.651
Concentrate (kg/d)	2.4	2.3	0.05	0.663
Total (kg/d)	4.6	4.5	0.07	0.751
DMI (% b. wt.)	3.3	3.1	0.09	0.689
DMI (g /kg $^{0.75}$/d)	112.7	108.2	4.15	0.870
Nutrient digestibility				
DM	60.2	61.9	0.43	<0.011
OM	62.3	63.5	0.41	<0.010
CP	65.8	66.1	0.47	0.619
EE	69.6	70.1	0.58	0.758
NDF	52.7	53.9	0.39	<0.013
ADF	47.3	48.6	0.37	<0.011
Cellulose	49.7	50.9	0.41	<0.012
Nutritive value				
DCP (%)	7.3	7.2	0.14	0.652
TDN (%)	60.1	61.9	0.39	<0.011
Plane of nutrition				
DCP intake				
g/d	335.8	324.1	14.25	0.893
g/kg body weight/d	2.4	2.2	0.08	0.619
g/kg w$^{0.75}$/d	8.3	7.8	0.17	0.693
TDN intake				
kg/d	2.8	2.8	0.03	0.689
g/kg body weight/d	19.9	19.3	1.05	0.704
g/kg w$^{0.75}$/d	70.1	66.9	2.13	0.738

NS, Non significant; SEM, Standard error of mean.

and results indicated that dietary supplementation of *Ficus hookeri* leaf meal as herbal feed additive did not have any effect on voluntary feed intake. Earlier it was also reported that supplementation of *Sapindus mukorosssi* leaves @ 3% in diet as herbal feed additive had no effect on daily feed intake in Rathi calves (Meel *et al*. 2015). Similarly, a non-significant effect of tropical tree leaves supplementation as herbal feed additive on dry matter intake (DMI, g/d) was reported in sheep (Malik *et al.* 2017). However, Chaturvedi *et al*. (2021) reported that daily dry matter intake becomes increased due to supplementation of herbal feed additive in growing Barbari kids reared under confined condition.

The digestibility of dry matter, organic matter, neutral detergent fibre, acid detergent fibre and cellulose improved (P<0.01) due to dietary supplementation of *Ficus hookeri* leaf meal as feed additive. However, digestibility of crude protein and ether extract was not influenced by dietary supplementation of *Ficus hookeri* leaf meal. In the present experiment, dry matter digestibility improved by 2.7% due to dietary supplementation of *Ficus hookeri* leaf meal as feed additive. Earlier it was reported that dry matter digestibility was increased by 4.8% due to dietary

Table 3. Effect of dietary Da qing shu (*Ficus hookeri*) leaf supplementation as herbal feed additive on blood metabolites, growth performance and feed conversion efficiency in growing cross-bred calves

Attribute	Level of *Ficus hookeri* leaf meal (%)		SEM	P value
	00 % (G1)	2.0 % (G2)		
Blood profile				
Plasma glucose (mg/dl)	60.5	63.8	0.39	<0.051
Plasma urea (mg/dl)	36.1	34.9	0.57	0.758
Serum total protein (g/dl)	6.9	7.2	0.11	0.716
Serum albumin (g/dl)	3.1	3.1	0.05	0.685
Serum globulin (g/dl)	3.8	4.1	0.08	0.638
Albumin:globulin ratio	0.81	0.76	0.04	0.692
Growth performance				
Initial body weight (kg)	92.1	91.7	1.29	0.683
Final body weight (kg)	166.7	175.3	2.06	<0.013
Total body weight gain (kg)	74.6	83.6	1.36	<0.011
Average daily body weight gain (g)	532.9	597.1	8.41	<0.011
Plane of nutrition during experimental period				
Total DM intake (kg)	534.9	526.4	21.52	0.895
Total DCP intake (kg)	39.1	38.4	2.91	0.683
Total TDN intake (kg)	321.5	333.7	18.73	0.738
Feed conversion efficiency				
DM intake (kg/kg gain)	7.2	6.3	0.04	<0.013
DCP intake (kg/kg gain)	524.1	459.3	11.75	<0.012
TDN intake (kg/kg gain)	4.3	3.99	0.03	<0.051
Feed efficiency (%)	13.9	15.8	0.04	<0.053

NS, Non significant; SEM, Standard error of mean.

supplementation of *Piper sarmentosum* leaf meal at 2.4 g/head/d in cattle in comparison to control (Cherdthong *et al.* 2019). This result could be attributed to protozoa, which are capable of ingesting fibrolytic bacteria; therefore suppressing these protozoa is expected to increase number of fibrolytic bacteria in the rumen, which can enhance feed digestibility in animals (Cherdthong *et al*. 2019). However, the present study could not confirm whether fibrolytic bacterial number enhances or not to improve feed digestion. Previous *in vitro* studies in our laboratory confirmed that inclusion of *Ficus hookeri* leaf meal in incubation media reduced rumen protozoal population (Choudhary 2016). Meel *et al*. (2015) reported that incorporation of *Sapindus mukorossi* (Reetha) leaf @ 3% in the diet as herbal feed additive improved the digestibility of nutrients in Rathi calves. Similarly, supplementation of *Moringa oleifera* leaves, improved nutrient digestibility as well as fermentation of rumen in dairy cows (Li *et al.* 2019). The use of 50 mg/kg of *Aloe vera*, *Azadirachta indica*, *Moringa oleifera*, *Jatropha curcas*, *Tithonia diversifolia* and *Carica papaya* extract to a forage-based diet reduced methane production while improving feed digestibility under *in vitro* system (Akanmu *et al.* 2020). Findings of the present experiment, e.g. digestibility of CP and EE was not influenced by the dietary supplementation of herbal feed additive is also similar to the earlier finding in growing kids in which CP and EE digestibility was not influenced by the dietary supplementation of mixture of herbal feed additive (Chaturvedi *et al*. 2021).

Digestible crude protein (DCP) content of ration was similar in G1 and G2 group. However, total digestible nutrients (TDN) content of the ration was higher (P<0.01) for *Ficus hookeri* leaf meal fed calves (G2). Higher TDN content in the ration of G2 group might be due to higher nutrient (DM, OM, NDF, ADF and cellulose) digestibility. Dietary supplementation of *Ficus hookeri* leaf meal as herbal feed additive had no effect on daily nutrient intake/ plane of nutrition of growing calves. Similar plane of nutrition of the two experimental groups, e.g. G1 and G2 in the present experiment was due to similar daily dry mater intake between the two experimental groups as well as slightly higher body weight of the calves in G2 group during digestion trial. Further, intake of DM, TDN and CP/DCP in calves under all two treatments was estimated adequate as the recommended requirements of NRC (2001) for growing calves with average daily body weight gain of about 600 g.

Blood profile, body weight changes and feed conversion efficiency: Blood glucose level was higher (P<0.05) in the *Ficus hookeri* leaf meal fed calves (G2). Dietary supplementation of *Ficus hookeri* leaf meal as feed additive had no influence on plasma urea, serum total protein and serum albumin concentration (Table 3). Higher blood glucose level in *Ficus hookeri* leaf meal fed calves might be due to higher nutrient digestibility and ruminal propionate production (Santra *et al.* 2013b).

Final body weight, average daily body weight gain and feed conversion efficiency were higher (P<0.01) for the

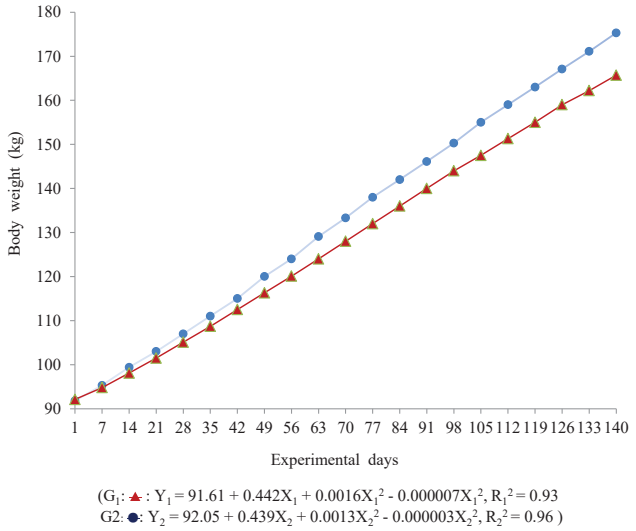

Fig. 1. Body weight changes of crossbred calves maintained on 50:50 paddy straw and concentrate based diet.

calves supplemented with *Ficus hookeri* leaf meal (G2) than the non supplemented calves (G1). Better growth rate and feed conversion efficiency in the *Ficus hookeri* leaf meal fed calves might be due to higher nutrient digestibility resulting in better utilization of dietary energy, leading to better body weight gain and feed conversion efficiency. Calves of G2 group started to gain in more body weight even after seven days of experimental feeding (Fig. 1). Nutrient intake, e.g. DM, DCP and TDN intake during total experimental period was similar among the calves of two experimental groups (G1 and G2). The calves of G1 and G2 group consumed 112.7 and 108.2 g DM, 8.3 and 7.8 g DCP, 70.1 and 66.9 g TDN per kg metabolic body size and had an average daily gain of 532.9 and 597.1 g, respectively. The calves of the present experiment meet the nutrient requirement of ICAR (2013) for DM, DCP and TDN. The presence of secondary metabolites such as flavonoids, saponins, tannins, etc. in plants, produce some biological activity in ruminant animals resulting in better body weight gain and feed conversion efficiency (Cherdthong *et al*. 2019). It was reported that *Ficus hookeri* leaf contains various phyto-chemicals like tannin, saponin, alkaloids, etc. (Bello *et al*. 2014) which might have reduced the ruminal methanogenesis and protozoal population resulting in a better body weight gain and feed conversion efficiency in the growing crossbred calves in the present experiment. In our laboratory, we observed inclusion of *Ficus hookeri* leaf meal in the culture media reduce ruminal methanogenesis and protozoal population *in vitro* (Shilpa 2016). Meel *et al*. (2015) reported that incorporation of *Sapindus mukorossi* (Reetha) leaf @ 3% in the diet as herbal feed additive improved the average daily gain and feed conversion efficiency in Rathi calves. It was observed that feeding of tanniniferous tropical tree leaves, e.g. *Ficus benghalensis*, *Artocarpus heterophyllus* and *Azadirachta indica* through concentrate mixture (10 parts, w/w basis, wheat bran in concentrate mixture replaced by these tree leaves) as a natural feed additive reduced ruminal methane production and improved body weight gain in sheep (Malik *et al*. 2017).

Feeding of sun dried *Ficus hookeri* leaf meal as herbal feed additive @ 2% of total diet/ration was effective for improving dry matter digestibility, body weight gain and feed conversion efficiency in growing crossbred calves. Thus, supplementation of Da qing shu (*Ficus hookeri)* leaf meal as herbal feed additive is recommended for manipulating rumen fermentation to improve growth performances of growing cattle.

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