

Effect of feeding maize silage containing Moringa (*Moringa oleifera*) leaves on growth, blood metabolites, serum antioxidant and coccidial egg count in Barbari goats under stall-fed condition

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ABSTRACT

Feeding cum growth trial was conducted on male Barbari goats to study the growth, blood metabolites, antioxidant and coccidial egg count on feeding of Maize silage having Moringa leaves. Eighteen male Barbari goats (Aged about 3 months and Avg B.Wt. 9.53 ± 0.42 kg) were divided in completely randomized design into (CRD) three groups of six each. Control group (Gr C) was fed with concentrate pellet, green and gram straw *ad lib*. Treatment groups (Gr M) were fed with concentrate pellet + maize silage *ad lib*.; Gr MM was fed with concentrate pellet + Maize –Moringa silage *ad lib*. Average daily gain (g/day) was highest in Gr MM (71.42) followed by Gr M (60.87) and Gr C (55.31). There was no significant difference in hematological and serum metabolites (Glucose, total protein, albumin, globulin, AG ratio) among groups. Serum cholesterol (mg/dl) was significantly lower in Gr MM as compared to Gr M and Gr C. There was improvement in the serum antioxidant activity of the goats fed with MM silage. Present study concluded that incorporation of *Moringa oleifera* in the maize silage improved the body weight gain with reduction in serum cholesterol in growing Barbari goats under stall feeding system.

Keywords: Blood metabolites, Goats, Growth, Maize, Moringa, Silage

Moringa oleifera is a rapidly growing soft wood plant that is mainly distributed in tropical and sub-tropical zones. In recent years, *M. oleifera* as animal feed has attracted interest owing to its rich nutrients and low antinutrient content (Makkar and Becker 1997). *M. oleifera* leaf contains high amounts of (23-30%) proteins, minerals, vitamins, and other secondary metabolites (Su and Chen 2020). Owing to high biomass production of these plants, it can serve as a good feed resource in the diet of ruminants. In recent years, Moringa leaves have been widely used as substitutes for traditional protein feeds for monogastric animals, ruminants and aquatic animals (Wu *et al.* 2013, Mahfuj *et al.* 2019). Majority of the study has been conducted on *M. oleifera* leaf meal in ruminants. Aregheore (2002) found that goats fed with fresh *M. oleifera* leaves at 20 and 50% as replacement for batiki grass had higher live-weight gain and higher digestibility of dry matter, crude protein, neutral detergent fibre, and organic matter than the control group. Kholif *et al.* (2018) reported that supplementation with *M. oleifera* leaf to replace 75% dry matter of berseem clover can improve feed utilization in Nubian goats. They also observed that Moringa diets increased serum total protein, albumin, and glucose levels but decreased cholesterol and

triglyceride levels. This study demonstrated that dietary supplementation with Moringa leaves is a potential strategy to improve meat quality. Moreover, *M. oleifera* leaf extract is a powerful ingredient for increasing feed intake, and the digestibility of dry matter, organic matter, and neutral detergent fiber and does not affect the digestibility of crude protein in Nubian goats (Kholif *et al.* 2019). As a nutrient source supplement to forage, *M. oleifera* leaf meal improves not only growth performance but also milk output and the quality of cows and goats (Sanchez *et al.* 2006, Babiker *et al.* 2017). Very limited studies are documented to utilize and preserve Moringa as silage and its utilization in the growing goat ration. Owing to these properties, Moringa was incorporated in preparation of Maize silage and it was evaluated in growing Barbari goats.

MATERIALS AND METHODS

Animal, feeding management and experimental design:

The present study was conducted at experimental shed of Animal Nutrition Management and Product Technology Division of ICAR-Central Institute for Research on Goats, Makhdoom, India. Geographically, the institute is located at 27° N latitude, and 78° N E longitude at 176 m above the sea level. Eighteen male Barbari goats of 3 months of age having average body weight 9.53 ± 0.42 kg were divided into three groups of six each as per CRD. Control group (Gr C) was fed with concentrate pellet, green and

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ad lib. gram straw. Treatment group (Gr M) was fed with concentrate pellet + maize silage *ad lib.*; Gr MM was fed with concentrate pellet + Maize–Moringa silage *ad lib.* Composition (on DM basis) of concentrate pellet was maize grain, 20%; barley grain, 20%; groundnut cake, 6%; soybean meal, 5%; mustard cake, 5%; guar korma, 5%; wheat bran, 15%; de-oiled rice bran, 14%; molasses, 7%; mineral mixture, 2% and Salt, 1%. All the experimental animals were fed with anticoccidial drug and treated for internal /external parasites before the start of experiment. The experiment was conducted as per the guidelines of Institutional Animal Ethics Committee.

The duration of the experimental feeding was 60 days. The animals were fed with concentrate pellet at 8 AM and after consumption of pellet they were fed with roughage portion of diet. Body weight of the animals was recorded at fortnight interval with the help of digital weighing balance.

Collection of blood/ fecal sample and its analysis: Blood was collected from each animal of all three groups at 0 and at the end of experimental feeding. About 10 ml blood was collected from all the experimental animals in the morning (before feeding) by jugular vein puncture. Out of 10 ml, 8 ml blood was taken into a clean and dry test tube and kept in slanting position for 45 min to separate serum; and rest 2 ml of blood was taken in eppendorf tube containing anticoagulant (EDTA) for hematological analysis. The blood samples were brought to the laboratory and centrifuged at 3000 rpm for 15 min to separate serum collected in small plastic eppendorf tubes (2 ml) and stored at -20°C for further analysis

The whole blood was analyzed for hematological parameters using hematology analyzer (Melet Schloesing Laboratories, France) standardized for goats as per manufacturer protocol.

The serum samples were analyzed for different biochemical constituent's, viz. glucose, total protein, albumin, cholesterol and triglycerides using diagnostic commercial kits (Autospan, Span diagnostic LTD.). Concentration of glucose, total protein, albumin, globulin, triglycerides, cholesterol, HDL, LDL were quantified using end-point assay using double beam spectrophotometer (UV-Vis spectrophotometer, Optizen, 3220UV, Mecasys.co. Ltd, Korea) as per manufacturers instruction. Superoxide dismutase (SOD) activity in serum samples was measured by an indirect method using SOD determination kit by SIGMA-ALDRICH Co. LLC. Blois (1958) method was used to measure free radical scavenging activity using 1, 1-diphenyl 2-picryl-hydrazyl (DPPH). Total antioxidant capacity was estimated using total antioxidant capacity Assay kit (catalog no MAK187) of Sigma-aldrich, USA.

Goats were dewormed with albendazole and toltazuril

(2 dose) and kept in cleaned disinfected house for about three weeks before the start of experimental feeding. Fecal samples were collected from rectum manually in aseptic condition on day 0 (before experimental feeding), and on 14, 28 and 48 days during course of experimental feeding.

Statistical analysis: The data collected during study were analyzed by ANOVA as per Snedecor and Cochran (1989) according to a complete randomized design using statistical software package (SPSS). Individual animals were considered as experimental units. The difference between means was significant at 95% level of significance $P < 0.05$.

RESULTS AND DISCUSSION

Chemical composition of feed and growth performance:

The nutrient composition of concentrate pellet, and silage used in feeding of goats is presented in Table 1. Dry matter and crude protein was higher in MM silage while organic matter, ether extract and fibre fractions (NDF, ADF, cellulose) were similar in both the silage.

The body weight changes of the goat demonstrated that there was no significant ($P < 0.05$) effect of silage feeding

Table 1. Chemical composition (% on dry matter basis) of concentrate pellet and silage fed during experimental trial

Component	Concentrate pellet	Maize silage	Maize-moringa silage
Organic matter	90.51	91.38	90.80
Crude protein	18.61	8.49	9.53
Total ash	5.49	8.62	9.20
Ether extract	3.71	1.64	1.96
Neural detergent fibre	14.78	62.07	58.88
Acid detergent fibre	2.75	40.13	36.17
Cellulose	2.58	6.11	4.46
Hemi-cellulose	12.03	21.94	22.70
Total carbohydrate	70.19	81.25	79.31

on body weight gain in different groups of goat (Table 2). However average daily gain (g/day) was highest in Gr MM (71.42) followed by Gr M (60.87) and Gr C (55.31). The initial body weight (kg) was 9.33, 9.48 and 9.82 which increased to 12.65, 13.13 and 14.10 in C, M and MM group of goats respectively.

Net gain in body weight (kg) was 3.32, 3.65 and 4.28 in C, M and MM group of goats respectively. Dry matter intake (g/day) was statistically similar in control group (414) and treatment group of goats (337 for M and 418 for MM). The difference in intake was mainly due to difference in intake of roughage portion of diet. However

Table 2. Effect of Moringa silage feeding on body weight gain in different group of goats

Attribute	Gr C	Gr M	Gr MM	Mean /SEM	P value
Initial body weight (kg)	9.33±0.69	9.48±0.73	9.82±0.88	9.53±0.42	0.910
Final body weight (kg)	12.65±1.16	13.13±1.19	14.10±1.19	13.25±0.56	0.607
ADG (g/day)	55.31±1.04	60.87±6.77	71.42±6.16	62.01±4.80	0.427

average daily intake (DM) from MM silage (250 g) was more as compared to M silage (168.83 g) in treatment group of goats during experimental trial. Incorporation of Moringa in the diet of sheep and goats has variable results on growth and nutrient balance. Aregheore (2002) found that goats fed with fresh *M. oleifera* leaves at 20 and 50% as replacement for batiki grass had higher live-weight gain and higher nutrients digestibility than the control group. Supplementation of 25, 50, 75, and 100% *M. oleifera* leaf meal in daily diets as substitutes for cotton seed meals for rams did not change body weight gain relative to the body weight gain in the control group (Adegun and Aye 2013). Similarly, the trial of feeding goats with *M. oleifera* leaves did not show obvious distinction in feed intake, rumination rate, and digestibility compared with those in *Leucaena leucocephala*, which has been used in livestock feed productions for a long time (Manh *et al.* 2005). Fadiyimu *et al.* (2010) reported a predominantly high trend in crude protein intake, dry matter and nutrient digestibility, nitrogen retention, and hematological profile gain in West African dwarf sheep fed with *Panicum maximum* meal with 25% *M. oleifera* leaves supplements. Kholif *et al.* (2018) also reported that supplementation with *M. oleifera* leaf to replace 75% dry matter of berseem clover can improve feed utilization in Nubian goats. However in present experiment, 25% of maize has been replaced with Moringa leaves in silage preparation. The difference in result might be due to difference in the level of Moringa incorporation and the type of ration.

Hematology and blood metabolites: There was no statistically significant difference between groups in the different hematological parameters such as total RBC,

WBC, thrombocytes count and differential WBC count (Table 3). Hematological parameters give an idea about the general wellbeing and impact of ration on overall health of animals. *M. oleifera* leaf is rich in essential amino acids and iron ions, which are key components for hemoglobin and myoglobin formation. However blood hemoglobin (g/dl) was 8.25 for Gr C, 8.83 for Gr M and 8.15 for Gr MM and no significant difference was reported. Broiler chickens fed with *M. oleifera* leaf meal had increased red blood cell counts, especially the groups supplied with 0.6% *M. oleifera* leaf powder in basal diet; the highest mean value of red blood cell count was recorded in these chickens (Makanjuola *et al.* 2014). Fadiyimu *et al.* (2010) also reported hematological profile gain in West African dwarf sheep fed with *Panicum maximum* meal with 25% *M. oleifera* leaves supplements. In our study, all the groups were getting balanced nutrients for better blood profile. No significant difference was reported in serum glucose, total protein, albumin, globulin and AG ratio (Table 3). Serum cholesterol (mg/dl) was significantly reduced (84.19) in Moring silage fed group (Gr MM) as compared to Gr M (129.12) and Gr C (127.24). There was significant ($P<0.05$) lowering of LDL and HDL in Moringa fed groups of goats with no difference on serum triglycerides, Zeng *et al.* (2018) also reported that *M. oleifera* leaf meal can completely replace maize silage in lactating dairy cows diet but did not affect dry matter intake, milk yield, or milk composition. However, cows fed with over 50% *M. oleifera* leaf meal showed low serum concentrations of total cholesterol, high-density lipoprotein cholesterol, and low density lipoprotein cholesterol and higher serum concentrations of urea than the control group. The lowering

Table 3. Effect of Moringa silage feeding on hematology and serum metabolites in different group of goats

Attribute	Group C		Gr M		Gr MM	
	0 Day	End day	0 Day	End day	0 Day	End day
WBC (m/mm ³)	14.40±1.18	16.62±0.85	14.04±0.48	14.83±0.71	12.99±1.49	13.21±1.21
RBC (m/mm ³)	16.01±0.35	15.29±0.77	14.67±0.61	16.36±0.48	14.02±0.50	15.53±0.48
Hct (%)	25.18±1.07	21.46±1.05	21.21±1.30	23.10±0.85	19.46±0.64	22.21±0.62
Hb (g/dl)	8.00±0.14	8.25±0.46	7.56±0.33	8.83±0.32	7.25±0.28	8.15±0.29
Mcv (fl)	15.16±0.37	14.1±0.30	14.4±0.46	14.15±0.24	13.93±0.29	14.35±0.20
Mch (fl)	5.06±0.06	5.35±0.08	5.11±0.04	5.33±0.07	5.13±0.04	5.20±0.05
Mchc (g/dl)	33.93±1.34	38.33±0.69	35.63±1.15	38.21±0.52	37.2±0.78	36.6±0.43
Lymphocyte (%)	56.93±3.81	56.16±3.29	56.65±7.16	52.66±2.07	46.26±2.84	57.16±2.79
Monocyte (%)	3.38±0.14	10.05±0.97	4.06±0.45	10.21±1.08	4.66±0.31	9.35±0.90
Granulocyte (%)	39.55±3.51	33.78±2.91	40.41±7.15	37.11±2.05	49.05±3.15	33.48±2.88
Glucose (mg/dl)	89.37±5.85	64.69±1.49	90.26±2.84	62.02±2.19	71.73±4.24	71.03±4.50
Total protein (g/dl)	5.94±0.44	6.46±0.32	5.47±0.51	6.22±0.26	6.60±0.55	6.57±0.48
Albumin (g/dl)	3.29±0.07	3.46±0.14	3.55±0.13	3.57±0.14	3.14±0.19	3.89±0.12
Globulin (g/dl)	2.64±0.24	2.99±0.15	2.32±0.23	2.65±0.66	3.12±0.55	3.05±0.18
AG ratio	1.45±0.27	1.26±0.21	1.28±0.11	1.44±0.20	1.36±0.43	1.30±0.07
Cholesterol (mg/dl)	131±2.02 ^a	127.24±3.33 ^a	127.33±4.27 ^a	129.12±3.65 ^a	125±4.44 ^a	84.19±2.22 ^b
Triglycerides (mg/dl)	87.90±3.30	71.79±1.82	84.49±2.67	76.17±2.33	77.14±3.62	74.55±3.05
LDL (mg/dl)	51.24±5.82 ^a	41.96±3.42 ^a	42.84±4.71 ^a	30.88±4.40 ^a	49.92±4.73 ^a	12.6±1.14 ^b
HDL (mg/dl)	68.73±5.40 ^a	67.77±1.75 ^a	70.63±2.91 ^a	68.71±2.58 ^a	61.61±2.30 ^a	43.64±3.52 ^b

^{a,b} Values with different superscript in a row differ significantly ($P<0.05$).

of serum cholesterol content might be due to different metabolites like flavonoids in the moringa leaves.

Serum antioxidant activity: Measurement of total antioxidant capacity (TAC) of serum samples is indicative of their ability to counteract oxidative stress induced damage in cells. TAC is used to provide insights into oxidative-stress of the animals. Serum antioxidant was measured as SOD activity, DPPH % inhibition and total antioxidant capacity (Trolox equivalents) in this study. There was no significant difference ($P < 0.05$) in the serum antioxidant activity in different group of goats (Table 4).

The DPPH is a stable free radical used to evaluate the ability of compounds to act as free radical scavengers

Table 4. Effect of Moringa silage feeding on antioxidant activity in different group of goats

Attribute	Group C	Gr M	Gr MM
SOD (U/ml)	69.63±1.46	77.75±1.01	72.05±1.45
DPPH (% inhibition)	51.12±0.68	48.83±1.55	51.06±1.63
Conc. of antioxidant (Trolox equivalents) (nmol/μl)	14.87±0.89	15.27±0.89	14.23±0.96

and to measure the antioxidant capacity of compounds. There was no significant difference in DPPH activity among the different groups, the mean values being 51.12, 48.83 and 51.06, respectively in Gr C, Gr M and Gr MM. Concentration of antioxidant (Trolox equivalents) (nmol/μl) was also similar among groups, being 14.87, 15.27 and 14.23 in Gr C, Gr M and Gr MM respectively. *M. oleifera* leaf supplements confer high antioxidant capacity due to its high concentration of vitamins, flavonoids, phenols, and carotenoids (Pakade *et al.* 2013). It is incorporated in silage formation but no significant difference was observed. This might be due to degradation of metabolites during anaerobic fermentation and silage preparation. However feeding of Moringa leaves supplements has been reported to provide high antioxidant capacity in goats' meat (Qwele *et al.* 2013).

Parasitic load: The mean FOC and FEC on day 0 was zero as no oocysts and nematode eggs were observed (Table 5). With the advancement of feeding, faecal oocysts were recorded on day 14, 28, 48 and faecal coccidian oocysts were observed. Analysis of faecal oocyst count (FOC) data revealed that mean of FOC in Gr C (15, 27.8) and Gr M (3, 41.75) was apparently more as compared to mean FOC values seen in Gr MM (4, 22.8), the animal group fed with maize-moringa silage. However, between groups FOC means were not significantly different.

There were some reports of anticoccidial activity of *Moringa* in Broilers. El banna *et al.* (2016) reported that inclusion of *Moringa oleifera* powder (0.5 and 1%) to broiler's feed were very efficacious and equal to diclazuril (1 ppm) in preventing symptoms of coccidiosis associated with experimental infection with mixed *Eimeria* species. However Moringa treatment did not affect *Coccidia* egg counts, FAMACHA score, PCV or body weight in Boer goats infected with gastrointestinal parasites (USDA). They concluded that Moringa supplementation modulates inflammatory gene expression, oxidative stress and parasite burden in a species-specific manner in ruminants through cell mediated immunity. Preliminary results from another independent small study in goats suggest that *Moringa* consumption had a positive effect on animals with low worm counts but there was no effect on animals with high-count *Haemonchus* infection.

Present study concluded that incorporation of *Moringa oleifera* at the rate of 25% in the maize silage improved the body weight gain with reduction in serum cholesterol in growing Barbari goats under stall feeding system.

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Table 5. Means of Coccidial oocysts count (OPG) in different group of goats

Day of fecal collection	Group C		Gr M		Gr MM	
	Log transformed mean	Geometric mean	Log transformed mean	Geometric mean	Log transformed mean	Geometric mean
0 day	4.60±0.00	0	4.60±0.00	0	4.60±0.00	0
14 day	4.70±0.079	2000	4.63±0.00	600	4.64±0.015	800
28 day	4.61±0.00	100	4.65±0.021	1000	4.64±0.02	800
48 day	4.79±0.079	4000	4.80±0.128	4400	4.75±0.099	3200

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