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Effect of Fibrolytic Enzyme on Nutrient Utilization and Rumen Fermentation Pattern in Sheep

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ABSTRACT

Effect of fibrolytic enzyme supplementation on nutrients utilization of sheep was studied. Twelve male Patanwadi sheep (12-15 month) were randomly divided in two groups of 6 each. Sheep were fed rations containing 50 % wheat straw and 50% compound concentrate mixture. A fibrolytic exogenous enzyme @ 0.025% was added to the ration of animal fed TMR. At the end of 14 weeks experimental period, animals were kept on digestion trial and three animals of each group were used for rumen fermentation analysis. There was no difference in final body weight between the groups due to enzyme treatment. There was non-significant difference on intake of dry matter and organic matter and also digestibility of DM, OM, CF and EE were not affected by treatment. However, the digestibility of CP and NFE were observed significantly ($p<0.05$) higher in treatment group as compared to control group. There was no significant effect on pH, TVFA, ammonia-N and NPN concentrations. However, the Total-N and protein-N concentration were significantly higher in treatment group as compared to control group. The daily feed cost was ₹ 6.04 and 5.90 under T_1 and T_2, respectively ($P>0.05$) which was somewhat lower in T_2 in comparison to T_1. It could be concluded that fibrolytic enzyme have no effect on feed intake, body wt gain, digestibility except protein utilization.

Keywords: Fibrolytic enzyme, patanwadi sheep, nutrient digestibility, roughage

In India ruminant feeding system relies mainly on fibrous crop residues. In crop residues the lignocellulose component is the potential source of energy but the utilization of such residues by ruminants as feedstuffs is limited due to lack of lignolytic activity of the rumen microbial population (Thakur *et al.* 2010). Large quantities of biologically active enzymes as animal feed additives are now produced at low cost since recent improvements in fermentation technology and biotechnology. It is acknowledged that enzyme preparations with specific activities can be used to drive specific metabolic and digestive processes in the gastrointestinal tract and may enhance natural digestive processes to improve the availability of nutrients and feed intake thereafter (McAllister *et al.* 2001 and Colombatto *et al.* 2003). The use of biotechnology such as exogenous fibrolytic enzymes (EFE) to enhance the quality and digestibility of fibrous forage is on the verge of delivering practical benefits to ruminant production systems. In this regard, cellulases and xylanases are respectively amongst the two major enzyme groups that are specified to break β1-4 linkages joining sugar molecules of cellulose and xylans found in plant cell wall components (Beauchemin*et al.* 2003). Keeping on

the above fact, the effect of supplementing exogenous fibrolytic enzymes on nutrients utilization of sheep was investigated.

MATERIALS AND METHODS

The present experiment was conducted on 12 Patanwadi adult sheep to know the effect of fibrolytic enzyme supplementation on nutrient utilization at Animal Nutrition Research Department, College of Veterinary Science and Animal Husbandry, Anand Agricultural University, Anand. Twelve male Patanwadi sheep (12-15 month) were randomly divided into two groups of 6 each. The experiment diets composed of 50 % wheat straw and 50% compound concentrate mixture. A fibrolytic exogenous enzyme @ 0.025% was added to the ration of animal fed TMR. Free choice of fresh water was offered to all the sheep. During the preliminary period, daily feed intake and body weight at weekly internal were recorded throughout of 15 weeks of experimental trial.

The digestion trial was conducted on all the sheep once at the end of experimental period to study the digestibility of nutrients. The collection period was of 5 days during which the representative samples of daily TMR fed, left over and output of faeces were collected and recorded every morning. Dried samples of feed, leftover and faeces of the 5 days collection period were pooled, ground to pass through a 1-mm screen and preserved for chemical analysis. The wet faeces preserved in acid were used for estimation of faecal N.

About 50 ml of rumen liquor was collected from each sheep using stomach tube and employing suction (Lane *et al.* 1968) once during the experiment. The samples were collected at 0 hr (pre feeding) and at 3 and 6 hrs post feeding to study progressive changes in the pH, total volatile fatty acids (TVFA) and various nitrogenous constituents of strained rumen liquor (SRL). The rumen liquor was immediately brought to the laboratory and strained through four layers of cheese cloth. The pH of SRL was determined using "Systronic" digital pH meter having glass and reference electrodes. The total volatile fatty acids were determined by Markham steam distillation method (AOAC, 1995). Ammonia nitrogen was estimated as per Conway (1957) method immediately from SRL. However, total-N was determined by Kjeldahl's method. Rest of the SRL was stored in glass bottles by adding mercuric chloride @ 0.5g/100 ml SRL (Rust *et al.* 1965) and preserved in refrigerator for further analysis. Non protein nitrogen was estimated from SRL following Kjeldahl's method after precipitating the protein with 20% trichloro acetic acid solution. However, protein nitrogen content was calculated by difference of total nitrogen and non-protein nitrogen.

Cost of feeding under both the treatments was calculated from the records of daily feed consumption and by considering the procurement cost of feeds used for feeding of experimental sheep. The data generated were analyzed statistically using independent t test as per Snedecor and Cochran (1994). Chemical composition of the experimental feeds is shown and Table 1.

Table 1: Proportions (%) of ingredients used to prepare TMR and chemical composition (% on DM basis) of TMR used under experiment

Chemical composition	TMR (50% WS + 50% CCM)
CP	09.68
EE	01.97
CF	28.03
NFE	47.39
Ash	12.93
Silica	04.70
Ca	00.56
P	00.85

WS = Wheat straw, CCM = Compound concentrate mixture.

RESULTS AND DISCUSSIONS

Live wt and feed intake

Final body weight of Patanwadi sheep during experimental period was 28.87 and 27.76 kg under T_1 and T_2, respectively and the differences are statistically non-significant (P>0.05). Non-significant difference in body weight (33.7 and 34.0 kg under control and treatment group, respectively) were also reported by Muwalla *et al.* (2007) in Awassi lambs fed high concentrate diet without and with fibrolytic enzyme. Higher body weight of goats fed total mixed ration (with 65:35; forage: concentrate ratio to which the enzyme was, or was not, added to the concentrate) was reported by Gonzalez-Garcia *et al.* (2008) with non-significant difference. Similar findings with non-significant effect on body weight was also reported by Awawdeh and Obeidat (2011) in lambs fed diets containing olive cake without and with fibrolytic enzyme. Final body weight was similar in growing Barki sheep fed ration containing 70% concentrate and 30% of either berseen hay, corn stalk or wheat straw and fibrolytic enzyme was added to the ration of animals reported by Gad *et al.* (2011). Mota *et al.* (2011) conducted a study to evaluate the effect of exogenous enzyme, glucoamylase from

Table 2: Effect of fibrolytic enzymes supplementation on nutrients intake, body weight, digestibility and rumen fermentation of Patanwadi sheep fed TMR

Parameter		Control	Treatment	P value
Nutrients intake				
DMI	(g/d)	747.48 ± 32.15	717.94±24.83	NS
	Kg/100kg	2.60 ± 0.03	2.61 ± 0.03	NS
	g/kg $W^{0.75}$	60.06 ± 0.28	59.35 ± 0.06	NS
OMI	g/d	617.82 ± 26.07	593.73 ± 19.74	NS
	Kg/100 kg	2.15 ± 0.02	2.14 ± 0.02	NS
	g/kg $W^{0.75}$	49.65 ± 0.23	49.13 ± 0.07	NS
CPI	g/d	72.28 ± 3.06	71.43 ± 2.39	NS
DCPI	g/d	57.79 ± 2.59	60.17 ± 2.76	NS
TDN	g/d	473.71 ± 17.21	446.31 ± 12.83	NS
B.W.	kg	28.87 ± 1.54	27.76 ± 1.21	NS
Rumen fermentation pattern				
pH		6.64 ± 0.11	6.57 ± 0.19	NS
VFA	mEq/L	118 ± 9.57	124 ± 11.88	NS
NH_3-N	mg/dl	9.24 ± 1.56	9.26 ± 1.09	NS
Total-N	mg/dl	82.48^{b} ± 16.89	88.36^{a} ± 15.15	0.05*
NPN	mg/dl	70.00 ± 20.58	69.69 ± 10.01	NS
Protein- N	mg/dl	14.03^{b} ± 2.57	18.42^{a} ± 5.36	0.05*
Digestibility of nutrients (%)				
DM		55.45 ± 1.25	57.13 ± 0.24	NS
OM		68.49 ± 0.10	68.26 ± 0.09	NS
CP		78.14^{b} ± 1.06	84.27^{a} ± 1.14	0.01**
EE		73.74 ± 0.69	73.37 ± 0.74	NS
CF		72.53 ± 0.47	70.55 ± 0.59	NS
NFE		62.25^{b} ± 0.47	64.99^{a} ± 0.58	0.05*

Means with different superscript in a row differ significantly *p<0.05, **p<0.01
NS=Non-significant, $W^{0.75}$ =Metabolic body weight, B.W. = Body weight

Aspergillusniger addition in finishing diets for lambs. The average daily gain (kg) not affected by the treatments.

The feed intake (g/d, % BW and g/kg W $^{0.75}$) between two groups was similar suggesting that the fibrolytic enzyme did not affect the palatability of diet (Table 2). Similar results of unchanged dry matter intake were also evident in the study by Torres *et al.* (2013). Singh and Das in 2009 reported (DMI) was 542 and 530.2 g/d in groups T1 and T2, respectively, which was similar between groups. Similar findings were observed in studies in which enzyme treated total mixed ration was fed to lambs by McAllister *et al.* (2001). No effect in feed intake was observed in Awassi lambs fed high concentrate diet without or with fibrolytic enzyme supplementation (Muwalla *et al.* 2007). Intake of CP, DCP and TDN was also similar in both the groups.

Rumen metabolites

The non significant differences were observed for average value of pH, TVFA and ammonia-N concentrations. However, the total-N and protein-N concentrations were significantly higher in treatment group as compared to control group (Table 2). Similarly, a significant difference (P<0.05) in protein N concentration was observed by Thirumalesh *et al.* (2003) in sheep fed with T_1 (control) containing 40% ground *bajra* straw and 60% concentrate mixture separately, average concentration of NPN was similar in both the groups. Pinos-Rodrriguez *et al.* (2002) reported that enzyme did not change pH, but total VFA and Nitrogen retained was higher in treated group than control. Titi and Tabbaa (2004) found that Enzyme-treated lambs retained more (P < 0.05) nitrogen in their bodies than those fed the control diet. Rojo *et al.* (2005) reported that Ruminal pH was increased in amylase treated group than control whereas total VFA concentration linearly reduced in amylase treated group with an average 55.20mmol/L than control (68.70 m mol/L). The supplementation with enzyme did not affect urinary excretion of purine derivatives, ruminal pH, or concentrations of NH_3-N and total VFA found by Giraldo *et al.* (2008). Bhasker *et al.* (2013) conducted total volatile fatty acids (p<0.01) and ammonia-N concentration was higher in enzyme supplemented group, while no effect was observed on dry matter intake, ruminal pH and total nitrogen concentration.

Digestibility of nutrients

The digestibility of DM was numerically higher but not significantly higher in treatment group as compared to control group. The enzyme had no effect on digestibility of OM, EE and CF. However, digestibility of CP and NFE was significantly higher in fibrolytic enzyme supplemented group. Dry matter, OM, CP, and NDF digestibilities were all unaffected by the enzyme inclusion were found by Muwalla *et al.* (2007) and Awawdeh and Obeidat (2011). Giraldo *et al.* (2008) reported Enzyme supplementation did not affect digestibility of any nutrient in Sheep. Dry matter digestibility increased in enzyme supplemented goats, while digestibilities of organic matter, protein and fibre fractions did not vary reported by Gonzalez *et al.* (2008). In contrast to this Titi and Tabbaa (2004) and Pinos-Rodrriguez *et al.* (2002) reported that Fibrolytic enzymes increased (P < 0.05) digestibility of DM, OM, CP, CF, and NDF than control group. Singh and Das (2009) reported that apparent digestibility coefficient of DM, OM, NDF and cellulose was significantly (P< 0.05) higher in animals fed on enzyme treated diet. Yang *et al.* (2001) who observed significantly higher digestibility of DM, OM and fibre fractions in lambs fed enzyme treated roughage. Singh and Das (2008) found a similar trend toward increased digestibility of nutrients when hay treated with fibrolytic enzymes were offered to goats. Enhanced TDN, CP digestibility with normal blood and rumen parameters in sheep were observed by Gomaa *et al.* (2012) after treating rations with exogenous enzymes.

Similarly daily feed cost was ₹ 6.04 and 5.90 under T_1 and T_2, respectively (P>0.05) which was somewhat lower in enzyme treated group compared to control group.

CONCLUSION

It may be concluded that application of fibrolytic enzyme have no effect on feed intake, growth performance, nutrient utilization except digestibility of CP, NFE, and rumen fermentation (Total-N and protein-N). Due to this concern, further studies needed to evaluate the factors that can affect the enzyme action to improve fiber digestion and animal performance.

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