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EFFECT OF ADMINISTRATION HIGH DOSES EXOGENOUS FIBROLYTIC ENZYME ON SOME RUMEN PARAMETERS IN LOCAL AWASSI RAMS

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ABSTRACT

This study carries out to examine the high doses effect of fibrolytic enzyme for improving the digestibility of sheep. Twelve Awassi sheep ram aged between 1.5-2 years were divided randomly into three groups (4 sheep each) housed in 3 experimental pens. First group (C) fed 3% body weight /day basal ration considered as control group, second group (T1) supplemented orally with 10g / head exogenous fibrolytic enzyme (xylanases, B-glucanases and cellulases) with 3% body weight /day basal ration, and third group (T2) supplemented orally with 20g/ head exogenous fibrolytic enzyme (xylanases, B-glucanases and cellulases) with 3% body weight /day basal ration. Rumen fluid were collected by stomach tube biweekly in order to detect the acidity, protozoa activity, Volatile fatty acids and NH3-N concentration in the rumen. The results showed a significant decrease ($P<0.05$) in the pH and a significant increase ($P<0.05$) in the volatile fatty acids (VFA) and nitrogen concentration while protozoa activity showed numerically increased with time progress in treated groups without significant differences between treated groups and control group. In conclusion, Awassi ram supplemented with exogenous fibrolytic enzyme had positive effect on Rumen performance with no significant deference's effect on Rumen traits measured in this experiment between (10g and 20g) of exogenous fibrolytic enzyme.

KEYWORDS: exogenous fibrolytic enzyme, protozoa activity, VFA, Rumen activity, rams.

INTRODUCTION

Biotechnology applications to Ruminant nutrition to improve the digestibility of structural carbohydrates by using exogenous fibrolytic enzymes in high quality forages (Colombatto *et al*., 2003; Dean *et al*., 2008). These enzymes are commercially produced from *Aspergillus niger, Aspergillus oryzae, Trichoderma viride* and *Trichoderma reesei* (Nadeau *et al*., 2000; McAllister *et al*., 2001). Some studies showed unpredictable results when these enzymes added to low quality forages (Avellaneda *et al*., 2009; Gallardo *et al*., 2010). Exogenous enzymes are used widely for removing anti-nutritional factors, and to increase the digestibility of existing nutrients, as well as to increase the activity of the endogenous enzymes (Classen *et al*., 1991; Bedford, 1993). Several studies have attempted to find the possible modes of action of these additives (Feng *et al*., 1996; Yang *et al*., 1998). Exogenous enzymes could utilize a number of effects, both on the gastrointestinal microflora and on the Ruminant animal itself. Hristov *et al*. (1996) and Gallardo *et al*. (2010) found that exogenous enzymes enhance fiber degradation by Ruminal microorganisms in vitro, and Yang *et al*. (1999) and Al-Deri (2014) in vivo. The exogenous enzyme increased feed digestion due to a number of contributing factors. Morgavi *et al*. (2000) confirmed synergism effect between exogenous enzymes and Ruminal enzymes like the net combined hydrolytic effect in the rumen was greater than that estimated from the individual activities. Therefore, the main aim of the inclusion of these enzymes is to increase the fibre digestibility of the diets fed, with the subsequent improvements in feed intake and animal production, amongst other. Preparations of enzymes that degrade cell walls (cellulases and xylanases) have the impending to hydrolyze forage fibre (Feng *et al*., 1996).

MATERIALS & METHODS

This study was conducted at the experimental station of College of Veterinary Medicine -Baghdad University (Abu Graib) using 12 Awassi sheep ram aged between 1.5-2 years, housed in 3 experimental pens with average body weights 46.50 Kg, the animals were randomly and equally divided into three groups (4 each). The experiment began from April and lasted to the end of June 2016, the animals fed concentrate diet (table, 1) 3% body weight /day and freely grazed for 3 hours/day on the college fields. The first group (C) received no supplementation and served as a control group, the second group (T1) supplemented orally with 10g / head exogenous fibrolytic enzymes (xylanases 14,000 U. Xyl, B-glucanases 12500 U.F. Pase and cellulases 50 U.F. Pase) daily. Third group (T2) supplemented orally with 20g / head exogenous fibrolytic enzymes (xylanases 28,000U.Xyl, B-glucanases 25000 U.F. Pase and cellulases 100 U.F. Pase) daily. The commercial Exogenous fibrolytic Enzyme (Safizym®-France) consisting of three exogenous fibrolytic enzymes (Xylanase (EC3.2.1.8): 1,400,000 U. Xyl/kg, B-glucanase (EC3.2.1.6) : 1,250,000 U.F. Pase/kg and Cellulase (EC3.2.1.4) : 5,000 U.F. Pase/kg was given orally to the animals one hour before the concentrate meal. Samples of rumen fluid were taken three hour after the concentrate meal by stomach tube biweekly intervals. The acidity of rumen fluid was detected immediately after obtained rumen fluid samples, according to (Hungat and El-Shazly (1965). The activity of the rumen protozoa was estimated by the relatively movement which have been refluxed its activity in more than three field at least (Joshi *et al*., 1977). Volatile fatty acids were detected by using Markham system, according to (Warner, 1964). While NH3-N concentration was determined by the spectrometer using the colorimeter with the NHCl solution as a standard (Feng and Gao, 1993).The data obtained from the performance test were analyzed by complete randomized design (CRD). Applied 5% level least significant difference (LSD) among different group (Steel and Torrie, 1980).

TABLE 1: The components of the concentrate diet

No	Nutritional substances	Percentages
2	Whole Barley	82 %
3	Soyabean meal	13.5 %
4	Calcium carbonate	1.5 %
5	NaCl	0.3 %
6	Primix	1 %

Crude Protein (CP) = 15.81 %, Energy = 1240 Kcal.

RESULTS & DISCUSSION

Acidity of rumen (pH)

The results of exogenous enzyme on the rumen acidity shown in the table (2). The pH of the both treated group were significantly lower than the control group from the second period and to the end of the study. Several researchers reported an effect similar to that observed in this study (Bueno *et al*., 2013 and Scholz *et al*., 2001), The pH values reflex carbohydrate fermentation in the rumen and absorption of the volatile fatty acids (Mutsvangwa, *et al*., 1992). The decrease in pH could be due to microbial Ruminal activity increases (Spoelstra *et al*., 1992). Bach *et al*. (2007) showed that pH value reduced and the total microorganisms increased by using probiotic as a feed additive. On other hand, the exogenous fibrinolytic microbial activity was most, improving the digestion of the cell walls in the diet, which in turn influence on the rumen acidity (Colombatto *et al.*, 2007).

TABLE 2: Effect of fibrolytic enzyme on the rumen acidity (pH) of local male rams (Means ±SE)

Groups Period	C	T1	T2
1	6.32±0.01	6.32±0.02	6.33±0.02
2	6.40±0.03 a	6.25±0.01 b	6.24±0.19 b
3	6.30±0.20 a	6.19±021 b	6.18±0.15 b
4	6.39±0.05 a	6.09±0.51 b	6.11±0.22 b

The different lowercase letters in a row refer to significant differences between different groups at (P<0.05).

Protozoa activity

Table (3) showed no significant differences between the treated groups and the control group. Although the treated groups T1 and T2 were higher in their protozoa activity mathematically compared with the control group along the studied period. The results designate that fed exogenous fibrolytic enzymes directly will increase the protozoal activity because they change Ruminal fermentation, intake, and digestibility of forages with different nutritive value (Pinos-Rodríguez *et al*., 2002). Also agreed with McSweeney and Mackie (2012) who found that protozoa might be responsible for 30–40% of overall fiber digestion under certain conditions. The increased butyric acid observed in this experiment promoted the release of simple carbohydrates such as glucose, possibly because of the presence of the *T.longibrachiatum* and *C. flavigena* fibrolytic enzymes, these enzymes encourage the populations of butyrate produced protozoa (Williams *et al*., 1986).

TABLE 3: Effect of fibrolytic enzyme on the protozoa activity of local male rams. (Means ±SE)

Groups period	C	T1	T2
1	1.56±0.62	1.75±0.10	1.62±0.07
2	1.75±0.00	2.37±0.16	2.50±0.10
3	1.93±0.06	2.62±0.07	2.43±0.06
4	1.93±0.11	2.68±0.06	2.68±0.14

The different lowercase letters in a row refer to significant differences between different groups at (P<0.05).

Volatile fatty acids (VFA)

Volatile fatty acids showed significant ($p<0.05$) increase from the second period of the study (195.0±3.39, 199.02± 1.10) for the T1 and T2 respectively compared with the (173.65± 3.37) of the control group. These results were take the same trend along the studied period (table, 4). The significant increase of the volatile fatty acids may due to enzymes applicable in improving Ruminal degradation of substrates DM and enzymes increased the true degradability of substrate DM and the creation of acetate, propionate, total volatile fatty acids and gas (Giraldo *et al*., 2008). While Yang *et al*. (2011) found that the proportion of volatile fatty acids was unchanged by the addition of elevated levels of EFE, and a decrease after applying EFE at 5 g /kg.

TABLE 4: Effect of fibrolytic enzyme on the volatile fatty acids (VFA) of local male rams (Means ±SE).

Groups period	C	T1	T2
1	175.75±1.86	179.57±0.42	179.55±1.10
2	173.65±3.37 b	195.00±3.39 a	199.02±0.63 a
3	173.62±2.48 b	205.45±2.45 a	208.65±1.19 a
4	1.77.60±0.92 b	222.37±2.73 a	225.37±2.43 a

The different lowercase letters in a row refer to significant differences between different groups at (P<0.05).

Ammonia - Nitrogen concentration (NH3-N)

Results showed that the treated groups T1 and T2 were significantly higher (p<0.05) than the control group from the second period and along the studied period (table, 5). The increase in the nitrogen concentration may because the enzyme increased apparent digestibility of CP, hemicellulose and NDF for alfalfa. Also the enzyme enhanced N balance because more N retained in lambs. The enzymes increase the digestion of DM and NDF, additional energy substrates are released, microbial protein synthesis improvement by reducing the concentration of N-NH 3 (Gado *et al*., 2011). While Giraldo *et al*. (2008) and Baah *et al*.(2005) found that ammonia nitrogen was not affected by the levels of EFE, consistent with other studies with sheep fed enzyme.

TABLE 5: Effect of fibrolytic enzyme on the Ammonia - Nitrogen concentration (NH3-N) of local male rams (Means ±SE).

Groups period	C	T1	T2
1	28.12±0.38	28.35±0.15	28.27±0.29
2	28.80±0.20 b	29.75±0.27 a	29.77±0.09 a
3	28.77±0.39 b	31.22±0.23 a	31.12±0.51 a
4	28.85±0.34 b	32.70±0.24 a	32.81±0.15 a

The different lowercase letters in a row refer to significant differences between different groups at (P<0.05)

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