Growth Performance and Nutrient Utilization in *Yankasa* Rams Fed Crop Residue Supplemented with Varying Levels of Xylanase and Glucanase Combinations

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Abstract

There is less information with regard the understanding of the use of exogenous enzymes in ruminant's rations in Nigeria. The study evaluated the effects of supplementing xylanase and glucanase combinations in rations of Yankasa rams on growth performance and nutrient utilization. Twenty Yankasa rams (average 23 kg) were used. Four treatments were evaluated: control (without enzyme combination); 50:50; 75:25 and 25:75 xylanase-glucanase combinations denoted as T_1, T_2, T_3, and T_4 respectively. The basal roughage was cowpea husk and sorghum husk. The feeding trial was conducted using completely randomize design. Growth performance and nutrient utilization parameters were measured. The growth parameters showed no significance ($p>0.05$) except average daily feed intake (ADFI). The ADFI (145.11 g/d) was increased in T_4 compared to other treatments. There were differences ($p<0.05$) with regard to nutrient intake and total digestibility coefficient except crude protein and nitrogen free extract digestibility ($p>0.05$). Xylanase-glucanase combination at 25:75 ratio increased DM intake by 211.90 g/d, increased DM digestibility by 17.73%, increased ADF digestibility by 1.17% and increased NDF digestibility by 1.02% compared to the control. The nitrogen balance in the body did not differ ($p>0.05$) with supplementation of 50:50 xylanase-glucanase combination compared to the control. The efficiency of nitrogen utilization did not differ between the control and 50:50 xylanase-glucanase combination. The combination of xylanase-glucanase at the ratio 25:75 respectively improved nutrient intake and digestibility but did not influence nitrogen utilization.

Keywords: *Digestibility; Exogenous fibrolytic enzymes; feed intake; nitrogen balance; weight gain.*

INTRODUCTION

The scarcity of feeds is one of the major constraints to livestock production in the northern region of Nigeria (Akinfemi et al., 2012). These are due to the shortage & cost of conventional feeds (consumed by humans) and the crop residues (consumed by a large number of animals) available are of poor nutritional status. These poor-quality roughages comprise a huge part of the feed readily available and its consumption results to a low plane of nutrition with attendant low productivity of the country's indigenous animals (Abdel Hameed et al., 2013). Even though these crop residues are available to the ruminants and are produced in large quantity annually (most of them comes as threshing by-product); little quantity is used as feeds. The rest are usually left on farm to rot or thrown away as waste (Millam et al., 2018).

These problems have directed research efforts towards harnessing and enhancing the utilization of these abundant arable crop residues. Therefore, the concept of matching available/abundant feed resources with the production of ruminants has consequently intensified research into the evaluation of crop residues for feeding livestock rather than discarding it as waste (Otaru et al., 2011). Similarly, since animal performance on these crop residues are poor, one of the ways to improve the quality of these crop residues is through enzyme supplementation (Beauchemin et al., 2019). Commercial fibrolytic enzymes such as xylanase and glucanase have been used in monogastric rations over the years. Few trials have tested its efficacy in ruminates especially in Nigeria (Millam et al., 2020). Such exogenous enzymes have been tried in other regions to have recorded improved weight gain, feed efficiency, digestibility and rumen degradability (Almaraz et al., 2016; Bhasker et al., 2013; Menezes-Blackburn & Greiner, 2015; Togtokhbayar et al., 2017; Torres et al., 2013; Yang et al., 2000). Therefore, the aim of this study was to determine whether the effects of using xylanase and glucanase commonly used in monogastric rations, could be used as feed enzyme additive for *Yankasa* rams to enhance growth performance and nutrient utilization.

MATERIALS AND METHODS

Experimental site

This experiment was carried out at the Small Ruminant Unit of Adamawa State University Teaching and Research Farm, Mubi. All research protocols and use of animals were approved by Adamawa State University, Institutional Animal Care and Ethics Committee (IACEC/ANP-A045/2020). It certifies that the procedures adhere to the International standards on animal use and practice.

Feed collection

The feed ingredients [Cowpea husk; sorghum husk; local brewer's residue (*burukutu* waste); maize offal; bone meal and salt] used for the experiment were obtained from *TIKE* cattle market, Mubi South Local Government Area, Adamawa State. The enzymes (xylanase and glucanase) were obtained from RONOZYME® MultiGrain (MG); DSM Nutritional Products Ltd, Switzerland – xylanase (Endo-1, 4-β-xylanase; EC 3.2.1.8) and glucanase [Endo-1, 3(4)-β-glucanase; EC 3.2.1.6 and Endo-1, 4-β-glucanase; EC 3.2.1.4].

Experimental animals and management

Twenty intact *Yankasa* rams (*Ovis aries*) aged 12-15 months with mean initial body weight of 22.7 ± 0.5 kg was used for this experiment. The rams were procured from *TIKE* cattle market. Prior to the commencement to the experiment, the animals were ear-tagged (for identification). The rams were given prophylactic treatments, consisting of intramuscular application of long-lasting antibiotics (Oxytetracycline LA®) and multivitamin at a dosage of 1 ml/10 kg body weight of the animals. They were drenched with 1 ml/10 kg body weight of albendazole and treated against ectoparasites with 0.5 ml/10 kg body weight of ivermectin

(Ivomec®). The rams were quarantined for 6 weeks. Adequate feed and clean fresh water were provided daily to the rams *ad libitum*.

Experimental treatment, housing, design and feeding of animals

The study evaluated the following dietary treatments: T_1, control (no supplementation of enzymes); T_2, with 50:50 ratio of xylanase-glucanase combination; T_3, with 75:25 ratio of xylanase-glucanase combination; T_4, with 25:75 ratio of xylanase-glucanase combination. The enzymes were incorporated into the rations at the rate of 10 g per 100 kg of feed (manufacturer's recommendation). The ration was formulated using computer method (least cost ration formulation software). The rations were mixed manually on a clean concrete floor using a shovel, then bagged and kept safe for the experiment. A sample from the formulated ration was collected, and its proximate compositions were determined using the procedures described by (AOAC, 2005). Neutral detergent fibre (NDF) and Acid detergent fibre (ADF) were determined by the methods of (van Soest et al., 1991).

Table 1: Dietary composition of the experimental rations

Parameters (%)	T_1	T_2	T_3	T_4
Cowpea husk	30.00	30.00	30.00	30.00
Sorghum husk	30.00	30.00	30.00	30.00
Maize offal	25.40	25.40	25.40	25.40
Local brewers' residue	12.60	12.60	12.60	12.60
Bone meal	1.50	1.50	1.50	1.50
Salt	0.50	0.50	0.50	0.50
Enzyme combination	0.00	0.01	0.01	0.01
Total	100.00	100.00	100.00	100.00
Chemical analysis (%)				
Energy (kcal/kg)	2994.30	2994.30	2994.30	2994.30
Dry matter	94.50	94.50	94.50	94.50
Crude protein	18.56	18.56	18.56	18.56
Ether extract	6.51	6.51	6.51	6.51
Ash	6.80	6.80	6.80	6.80
Nitrogen free extract	50.13	50.13	50.13	50.13
Neutral detergent fibre	51.43	51.43	51.43	51.43
Acid detergent fibre	46.55	46.55	46.55	46.55

Enzyme combination: the combination of both xylanase and glucanase at 50:50, 75:25 and 25:75 levels of inclusion to makeup 10 g.

The experimental animals were housed in a well ventilated, individual enclosure (2×2 m dimension) with corrugated iron roof, concrete floors and equipped with individual feeders and water troughs. These stalls were washed properly and disinfected a week before the commencement of the feeding trial. The rams were weighed using WeiHeng (WH-A series) potable electronic scale (WH-A08) and randomly assigned to the four dietary treatments with five rams per treatment in a completely randomized design. The rams were fed at 3% body weight individually throughout the feeding trial. The feed offered was adjusted at regular intervals of two weeks along with changes in weight. Clean fresh water was provided to the rams *ad libitum* throughout the period of the feeding trial which lasted for 12 weeks. The total ration for a day was separated into two portions of equal weight and supplied to the animals at 8:00 h and 14:00 h.

Data collection

The initial wight was taken at the beginning of the trial and subsequent weight of the rams was recorded fortnightly. Feed offered and left over was measured daily using electronic kitchen scale (WH-B05). The leftover was deduced from the feed offered to compute the daily feed intake continuously till the end of the feeding trial.

For the digestibility trial, the rams were housed in individual metabolic cages according to the procedures of Osuji et al. (1993). The metabolic cages were designed for separate collection of faeces and urine. Twenty-one days was used as adjustment period on the crates, while seven days was for the collection of faeces and urine. Daily faeces and urine voided out by each ram was collected and measured. At the end of the collection period, the total faeces were then bulked and representative aliquot (10%) sample was collected for chemical analysis. Total urine collection was made over urine container acidified with sulphuric acid; 10 ml 0.1N H_2SO_4 (Osuji et al., 1993). This was to avoid nitrogen loss. The collected urine was measured and 20 ml aliquot was sampled for nitrogen analysis. The nitrogen was analysed using the Kjeldahl method of nitrogen analysis (AOAC, 2005).

Growth performance, nutrient intakes and digestibility coefficient of the experimental rations were calculated using the methods described by (McDonald et al., 2010). Nitrogen retention was computed as the difference between nitrogen intake (nitrogen in feed) and nitrogen losses (nitrogen in urine and faeces) while nitrogen as percentage of intakes was calculated from the nitrogen retention/balance which was expressed as a percentage of nitrogen intake.

Statistical analysis

All statistical procedure were carried out and analysed according to PROC GLM procedures of (SAS, 2002). The effect of treatment means was tested at probability level of 95% ($p<0.05$) and significant effects were compared using Duncan Multiple Range Test.

RESULTS AND DISCUSSION

Growth performance of *Yankasa* rams

It was observed that there was no statistical variation for growth parameters among the experimental treatments (Table 2: $p>0.05$) except for the average daily feed intake ($p<0.05$).

Table 2: Growth Performance in *Yankasa* rams fed experimental ration

Parameters	T_1	T_2	T_3	T_4	SEM
Initial weight (kg)	23.09	22.90	22.57	22.22	0.59
Final weight (kg)	24.87	24.43	24.37	24.02	0.55
Weight gain (kg)	1.78	1.53	1.80	1.80	0.21
Average daily weight gain (g/d)	21.19	18.21	21.43	21.55	1.85
Feed intake (kg)	11.70	11.06	11.22	12.19	0.96
Average daily feed intake (g/d)	139.29^{ab}	131.68^{b}	133.52^{b}	145.11^{a}	3.63
Feed conversion ratio	6.57	7.23	6.23	6.73	0.60

^{ab}: Means with different superscript are significantly different, SEM: standard error of means

Higher average daily feed intake (145.11 g/d) was observed for T_4 compared to other treatments. This may be attributed to the enzyme activity (25:75 xylanase-glucanase combination) which was said to improve the palatability of the ration (Beauchemin et al., 2003). It may also be as a result of acceptably due to animal difference (Millam, 2016). The animal difference refers to some animals accepting to eat particularly more than the other animals especially when crop residues are involved. The enzyme activity is usually a function of the rations post-ingestive feedback manifested after consumption (Menezes-Blackburn & Greiner, 2015) in which digestibility is increased to reduced gut fill and trigger the consumption of more feed. This result was similar with those reported by Salem *et al.* (2011) who fed wheat straw supplemented with an enzyme to sheep. Also, some authors reported that a non-significant effect in feed intake when lambs ration was supplemented with exogenous enzymes (Almaraz et al., 2016).

Nutrient intakes of *Yankasa* rams

There were differences ($p<0.05$) among treatments with regards to all parameters measured (dry matter intake, DMI; crude protein intake, CPI; nitrogen free extract intake, NFEI; acid detergent fibre intake, ADFI and neutral detergent fibre intake, NDFI) for the nutrient intake (Table 3).

Table 3: Nutrient intakes in *Yankasa* rams fed experimental ration

Parameters (g/d)	T_1	T_2	T_3	T_4	SEM
Dry matter intake	959.60^b	923.00^b	916.20^b	1171.50^a	89.77
Crude protein intake	188.46^b	181.29^b	179.95^b	230.09^a	17.63
Nitrogen free extract intake	509.00^b	489.60^b	486.00^b	621.50^a	47.62
Neutral detergent fibre intake	522.20^b	502.30^b	498.60^b	637.60^a	48.85
Acid detergent fibre intake	472.70^b	454.70^b	451.30^b	577.10^a	44.22

^{ab}: Means with different superscript are significantly different, SEM: standard error of means

The values for T_4 (DMI, 1,171.50 g; CPI, 230.09 g; NFEI, 621.50 g; NDFI, 637.60 g, ADFI, 577.10 g) was observed to be higher statistically ($p<0.05$) for all the parameters compared to other treatments. The increased nutrient intake is an important factor for improved productivity in and utilization of feed by ruminants (Jiwuba et al., 2016) which may be associated with some factors such as the palatability of the feed, reduced fibre fraction, etc. (Rahman et al., 2018). These translate to the reason which may have brought about the outcome of these results. High feed intake as observed with the rams receiving 25:75 xylanase-glucanase combination was an indicator that the rams ate the ration due to the taste or smell (TheCattleSite, 2019) and consequently influence the prehension and increased intake (Table 2; average daily feed intake) frequency of the rams (Yahya et al., 2020). Therefore, the improvement or increased nutrient intake in a broad sense may be due to the ration having less lignified cell wall tissues as a result of hydrolysis brought about by enzyme supplementation (Beauchemin et al., 2019). This decreases the ration retention time in the rumen (Wadhwa & Bakshi, 2013). The results obtained here differ from those reported by several authors who studied the use of various fibrolytic enzyme complex and discovered that its supplementation did not affect the nutrient intake in sheep (Bueno et al., 2013; Torres et al., 2013; Varlyakov et al., 2010).

Digestibility coefficient of *Yankasa* rams

Statistical influence ($p<0.05$) was observed in all parameters measured for digestibility coefficient (Table 4) except crude protein and nitrogen free extract ($p>0.05$).

Table 4: Nutrient digestibility of experimental rations in *Yankasa* rams

Parameters (%)	T_1	T_2	T_3	T_4	SEM
Dry matter digestibility	55.28^b	60.53^b	62.28^b	73.01^a	5.16
Crude protein digestibility	94.04	94.85	95.75	95.80	2.15
Ether extract digestibility	97.68^a	96.96^b	97.41^{ab}	97.95^a	0.25
Ash retention	66.59^a	56.83^b	61.31^{ab}	68.76^a	4.55
Nitrogen free extract digestibility	75.85	76.06	75.75	76.65	0.86
Acid detergent fibre digestibility	64.77^c	65.14^b	65.09^{bc}	65.94^a	0.16
Neutral detergent fibre digestibility	78.80^c	79.39^b	78.63^c	79.82^a	0.20

^{abc}: Means of different superscript are significantly different, SEM: standard error of means

Treatment T_4 was observed to have improved the dry matter digestibility, DMD (73.01%); acid detergent fibre digestibility, ADFD (65.94%) and neutral detergent fibre digestibility, NDFD (79.82%) compared to other treatments. Improved, DMD; ADFD; and NDFD observed in this study might be as a result of enzyme supplementation (Beauchemin et al., 2019). This is an indication that the use of 25:75 xylanase-glucanase combination in the ration was successful in improving the digestibility of the crop residues within the gut of the rams. The results were consistent with the works of various authors who reported that exogenous enzymes supplementation in fibrous ration increased fibre digestibility in sheep (Meale et al., 2014; Salem et al., 2015; Sujani & Seresinhe, 2015; Valdes et al., 2015). These positive improvements associated with enzyme combination may have resulted from enhanced colonization of feed by increasing the number of ruminal fibrolytic microbes (Morgavi et al., 2000); increased non-fibrolytic microbes (Colombatto et al., 2003); increased rate of fibre degradation in the rumen (Valdes et al., 2015); increased rumen microbial protein synthesis (Salem et al., 2015) and increased total tract digestibility (Gado et al., 2011). However, the increased digestion of the fibre fraction (ADF and NDF) in the 25:75 xylanase-glucanase combination treatment compared to the control may also be related to reduce digesta viscosity (Hristov et al., 2000); altered ruminal fermentation (Gado et al., 2011) or reduction of rumen physical fill over time which will consequently increase the nutrient intake (Table 3; Morgavi et al., 2000).

Nitrogen balance of *Yankasa* rams

The results record no statistical ($p<0.05$) difference between treatments for nitrogen intake and urine nitrogen (Table 5). Nitrogen losses were relatively higher in T_4 (0.61 g/d), it might be attributed to higher loss of nitrogen in the faeces. Nitrogen obtained from the ration consumed by an animal is lost through urine (about 90%) and through faeces (about 10%). In a situation where the losses in faeces are high, it may lead to high nitrogen loss (Rufino et al., 2016).

Table 5: Nitrogen balance in *Yankasa* rams fed experimental rations

Parameters (g/d)	T_1	T_2	T_3	T_4	SEM
Nitrogen intake	2.97	2.97	2.97	2.97	0.00
Faecal nitrogen	0.21^b	0.22^b	0.30^a	0.31^a	0.01
Urine nitrogen	0.29	0.30	0.30	0.30	0.01
Nitrogen losses	0.50^c	0.51^b	0.60^a	0.61^a	0.01
Nitrogen absorbed	2.76^a	2.76^a	2.67^b	2.66^b	0.01
Nitrogen retained/balance	2.47^a	2.46^a	2.37^b	2.36^b	0.01
Nitrogen balance as % of (%)	83.27^a	82.92^a	79.66^b	79.44^b	0.41

^{ab}: Means of difference superscript are significantly different, SEM: standard error of means,

Literatures point out that the major reason which affects the loss of nitrogen through faeces may be the forage-concentrate ratios, since a high level of concentrate in the ration may result in an increased rate of passage which consequently increases nitrogen loss through the activities of the microbes (de Almeida et al., 2015; Elamin et al., 2012; McDonald et al., 2010). The absence of effects may also be due to the fact of the experimental ration been iso-nitrogenous (Table 1; crude protein). The nutritional demands of ruminants highlight the synchronization between protein and dietary carbohydrates in the rumen to maximize microbial synthesis, thereby reducing nitrogen loss through urine (Bastos et al., 2014).

The higher nitrogen absorbed (2.76 g/d for both T_1 and T_2) and nitrogen retained/balance (T_1, 2.47 g/d; T_2, 2.46 g/d) was not influenced ($p<0.05$) by supplementing the ration with xylanase-glucanase combination at 50:50 ratio because they are statistically same with the control. Nitrogen retention is often referred as a good method of estimating the quantity of nitrogen available for body tissue deposition (Bastos et al., 2014). Therefore, the results indicates that the rations were able to present adequate levels of nitrogen (Table 1; crude protein). Increased nitrogen retained/balance is associated with higher urea production in the liver and optimum excretion in the urine (Endo et al., 2015).

The nitrogen balance as percentage of intakes (T_2, 82.92%) otherwise known as efficiency of nitrogen utilization was not influenced ($p<0.05$) by xylanase-glucanase supplementation at 50:50 ratio because they are statistically same with the control. Thus, the positive nitrogen balance as percentage of intakes observed in all the treatments indicates that there was no excessive loss of nitrogenous compounds during the trial. This confirms that the digestible fraction of protein in the ration was adsorbed efficiently by the animals (Jetana et al., 2010). Also, this positive nitrogen balance is an indication that the protein and energy demands (Table 1; crude protein and energy) of the rams were satisfied most likely and there was protein retention in the body of the rams for tissue growth and replacements, thereby avoiding weight loss (Pereira et al., 2012).

Conclusions

Based on the results obtained in this study, it could be concluded that supplementation of xylanase-glucanase combination at 25:75 ratio improved the nutrient intake and nutrient digestibility of *Yankasa* rams. Hence, recommended as suitable catalyst for fibre hydrolysis of high fibre rations in growing *Yankasa* rams and small ruminants at large.

Acknowledgments

The authors thank the Animal Nutrition Laboratory of the Department of Animal Production, Adamawa State University-Mubi, and its technician – Mr. Yahaya Baba Kiri for carrying out the chemical analysis reported in this experiment.

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