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INTESTINAL MORPHOLOGY OF GROWING RABBITS (ORYCTOLAGUS CUNICULLUS) OFFERED CORN COB-BASED DIET SUPPLEMENTED WITH FEED ADDITIVES

T. A. ADEOSUN[1]*, A. O. GANIYU[2], R. B. FATAI[3], V. A. ADEPEGBA[4], J. B. ADEOGUN[5] AND O. A. ABU[6]

[1&2]*Federal College of Education (Technical), Bichi, Department of Agricultural Education, Kano State, Nigeria.* [3]*Federal College of Education, Kano, Department of Agricultural Education, Kano State, Nigeria.* [4, 5 & 6]*University of Ibadan, Animal Science Department, Oyo State, Nigeria*

ABSTRACT

A ten-week feeding trial was carried out to investigate the effects of corn cob-based diet supplemented with feed additives on intestinal morphology of growing rabbits. Forty eight growing rabbit bucks of mixed breeds (New Zealand White x Chinchilla), weighing 642.01±4.99g were randomly allocated to four dietary treatments at three replicates of

INTRODUCTION

The level at which Nigerians consume protein in terms of quantity and quality goes a long way in describing their state of nutrition (Taiwo *et al.*, 2005). Meat consumption in Nigeria reported as 2 kg/person/year is still below the average of 8 kg/person/year in ECOWAS region (Benard *et al.*, 2010). Some of the reasons as adduced by Alikwe *et al.* (2014) responsible for low consumption of animal protein in the tropics could be due to low productivity of the animals arising mainly from lack of good management in the livestock industry, very bad genetic make-up of the local breeds of animals, inadequate services from the veterinary officers, expensive nature of feed, drugs and very bad quality of most of our indigenous feedstuff. Although proteins can as well be sourced from plants, the

four rabbits each in a completely randomized design. Diet 1 ((Basal, corn cob-based diet with no additive), diet 2 [Basal diet + Probiotic (Bactofort® at 500g/ton)], diet 3 {Basal diet + Exogenous Enzyme (Cellulase at 500g/ton)} and diet 4 {Basal diet + Symbiotic (Bactofort® at 500g/ton + Cellulase at 500g/ton)}. While there was no significant difference in villus width, and ratio of villus height to crypt depth across the treatments in the duodenum, the observed values for villus height, crypt depth and muscle thickness followed similar pattern with the values recorded in all the treatments fed supplemented diets higher (P<0.05) than the control. Duodenal crypt width was significantly higher (P<0.05) in the groups fed probiotic, and probiotic + enzyme treated diets than control, the value obtained for enzyme treated group was however similar to control. In the ileum, diet supplementation with feed additives did not significantly influence the observed values for crypt depth, crypt width, and ratio of villus heigjht to crypt depth. Meanwhile, the results showed that corn cob-based diet supplemented with fed additives significantly (P<0.05) improved villus height and muscle thickness. Ileal villus width was statistically similar (P>0.05) between the treatments fed probiotic, and probiotic + enzyme treated diets and higher than control, while the observed value for enzyme treated group was similar (P>0.05) to control. It was therefore concluded that supplementing corn cob-based diet with feed additives (Bactofort and cellulase) either singly or combined improved intestinal morphology in growing rabbits.

Keywords: *Intestinal morphology, corn cob, feed additives, growing rabbits*

place of animal protein remains significant, owing to its balanced amino acid profile (Ogunsipe *et al*., 2014). To therefore bridge the gap created by lack of adequate animal protein consumption in Nigerians' diets, there should be a paradigm shift to the production of animal like rabbit which is well known to be easily managed. Commercial or backyard rabbitry in Nigeria is however faced with problems ranging from expensive nature of conventional feed ingredients, dry season feeding problem, enteritis (Rabie *et al*., 2011) due to inadequate fibre source in feed prevalent most especially among weaners. As a result of these, the use of alternative or non-conventional feedstuffs as advocated for by Adedire *et al.* (2012) is paramount in sustainability of commercial rabbitry. Cereal crop residue like corn cob is produced in large quantity in Nigeria, and always available all year round.

Therefore, if corn cob can be fed to rabbits to serve as fibre source, replacing the more expensive conventional fibre sources already in use, it could lead to reduction in feeding cost in rabbit production. According to Bawa *et al.* (2008), rabbits come first among non-ruminants in utilising coarse materials simply because their gastro intestinal tracts are well adapted to consumption of feedstuffs relatively rich in fibre. Also, high fibre materials which pose a serious challenge in terms of utilisation when fed to poultry are found to be beneficial to rabbits for preventing enteritis. Gidenne and Lebas (2002) opined that rabbit digestive physiology is quite accustomed to taking much of plant cell walls and that the favourable roles of less-digested fibre (e.g lignin and cellulose) on reducing the rate of gastro intestinal maladies in rabbits cannot be over-emphasized. Meanwhile, incorporating crop residues in animal ration is however, not without some major limitations like reduction in nutrients digestibility and poor nutritive value (Adeniji and Adewole, 2015). This could be as a result of very bulky crop residues, low in digestible nutrients and highly fibrous nature of these residues (Idachaba *et al*., 2015). Despite the adaptation of rabbits gastro-intestinal tract to feedstuffs rich in fibre like cereal crop residues, there is still need to boost the nutrient utilisation for optimum performance and feed cost efficiency. As a way out to all these problems, researchers like Buba and Shehu (2018) and Attia *et al.* (2012) reported significant improvement in intestinal morphology of chickens and rabbits when fed diets supplemented with feed additives. It is against this background that this study was designed to evaluate the intestinal morphology of growing rabbits fed corn cob-based diet supplemented with probiotic (Bactofort®) and exogenous enzymes (cellulase).

Materials and Methods

Experimental Site

The experiment took place at the Rabbitry Unit, Teaching and Research Farm, University of Ibadan, Nigeria. It is situated in the humid forest zone of South-West, Nigeria on latitude 7°7'N and longitude 3°5' to 3°36'E with mean annual

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temperature of 25°C - 30°C, relative humidity of 65% - 84%, average annual rainfall of about 1250mm and altitude between 200m and 300m above sea level.

Sources of Probiotics (Bactofort®) and Exogenous Enzymes (Cellulase)

The probiotic used was commercial Bactofort®, containing: *Lactobacillus acidophilus* (77 x 109 cfu/kg), *Enterococcus faecium* (44 x 109 cfu/kg), *Saccharomyces cerevisiae* (5,000 x 109 cells/kg) and *Bacillus subtilis* (2.2 x 109cfu/kg). It is manufactured by Biofeed Technology Inc., Brossand, QC, Canada, and mixed at the rate of 0.5g/kg according to manufacturer's recommendation. Cellulase is manufactured by Co-Suppliers Ltd, China. Cellulase system according to the manufacturer consists of three major components: endoglucanases (endo-1,4-β-D-glucanases), cellobiohydrolases (exo-1,4-β-D-glucanases), and β-glucosidases (1,4-β-D-glucosidase). It was as well mixed at the rate of 0.5g/kg according to manufacturer's recommendation.

Experimental Animals and Management

Forty eight growing rabbit bucks of mixed breeds (New Zealand White x Chinchilla), weighing 642.01±4.99g were used for the experiment. The rabbits were procured from a reputable farm in Abeokuta, Ogun State, Nigeria. On arrival, the animals were dewormed with Banmith. They were acclimatised for the first two weeks in the experimental site. During this period, control diet was slowly introduced to the rabbits to take the place of the diet the animals were being fed before. This is to ensure that there was no stomach upset in rabbit that could lead to enteritis and some other related problems. After the acclimatisation period, the rabbits were allotted to four dietary treatments. 12 rabbits were in each treatment, and there were three replications in a treatment at four rabbits per replicate arranged in a completely randomised design. Clean water was supplied *ad libitum*.

Experimental diets

Four experimental diets were formulated such that the control (basal diet) contained no supplementation. Diets 2, 3 and 4 were supplemented with bactofort, cellulase, and bactofort + cellulase, respectively. The ingredients composition of the basal diet is shown in Table 1 below. The experimental diets layout is as follows:

Diet 1: Basal diet

TIJSRAT
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Diet 2: Basal diet + Bactofort®
Diet 3: Basal diet + Cellulase
Diet 4: Basal diet + Bactofort® + Cellulase

Table 3.4: Gross composition of experimental diets

Ingredients	Basal Diet
Maize	20
Maize offal	10
Corn cob	20
Soyabean meal	20
Palm kernel cake	21.5
Others*	8.5
Total	100
Calculated nutrient composition:	
Dry matter	85.62
Crude protein	16.34
Crude fibre	12.11
Ether Extract	3.70
Ash	3.60
Nitrogen Free Extract**	49.87
Neutral Detergent Fibre	36.77
Acid Detergent Fibre	17.63
Acid Detergent Lignin	5.67
Hemicellulose***	19.14
ME (kcal/g)****	2.65

*Others: Cassava Flour = 5%, DCP = 2%, Oyster Shell = 1%, Table Salt = 0.25%, Premix = 0.25% **Calculated as 100 – (% moisture + % CP + % CF + % EE + % Ash) ***Calculated as %NDF - %ADF, ****Calculated using Pauzenga Formula, ME = Metabolizable Energy

Data Collection

Intestinal morphology

At the end of the experiment (70 days feeding trial), twenty four rabbits of similar body weight to the group average were selected from the treatments (2 rabbits per replicate) for this analysis. They were weighed and sacrificed by severing the jugular vein. They were then thoroughly bled before taking the gut samples. Intestinal samples of 2cm each was taken from two different segments (duodenum and ileum) of the intestine. Duodenum was cut 10cm away from the pyloric junction, while ileum was cut 10cm away from the ileo-caecal junction of the intestine. Thereafter, 10% neutral formalin was used to fix the specimens for 24-48 hours. Running tap water was then used to wash the fixed specimens, the specimens were then dehydrated in ascending grades of alcohol, cleared, embedded in paraffin wax-blocks, cut into thin sections (7-10 μm) and stained by haematoxylin and eosin. Microscope was finally used to examine the stained sections in measuring villus height and width, crypt depth and width, and muscle thickness, as described by (Abdel-Khalek *et al*., 2011).

Results

Table 2 below shows the intestinal morphology of growing rabbits offered diets fortified with feed additives. In the duodenum, no significant difference appeared in villus height among the treatments administered diets supplemented with feed additives. However, villus height was observed to be lower ($P<0.05$) in the control by 20.20%, 16.67% and 18.29% than the values obtained for probiotic, enzyme, and probiotic + enzyme groups respectively. No significant difference existed in the values recorded for villus width across the treatments. Crypt depth appeared in the same trend observed in villus height, having the value obtained in control lower ($P<0.05$) by 21.02%, 16.41% and 22.33% than the obtained values for probiotic, enzyme, and probiotic + enzyme groups respectively. Crypt width was similar ($P>0.05$) between probiotic and probiotic + enzyme groups, but higher ($P<0.05$) by 17.89% and 20.77% respectively than the value observed for control. Muscle thickness was also similar among the treatments offered feed additives-fortified diets. The treatments fed probiotic, enzyme, and probiotic + enzyme fortified diets were however found higher ($P<0.05$) in muscle thickness by 16.35%, 13.89% and 14.74% than control respectively. No significant difference occured in villus height ratio crypt depth in the duodenum.

In the ileum, villus height was lower ($P<0.05$) in control by 33.33%, 24.44% and 29.58% than the groups fed probiotic, enzyme, and probiotic + enzyme respectively; while no significant difference existed among the treatments administered diets supplemented with feed additives. No significant difference existed in villus width among the treatments offered diets supplemented with feed additives, but treatments fed with probiotic and probiotic + enzyme treated diets were higher (P<0.05) by 24.27% and 21.94% than control respectively. There was no significant difference in crypt depth and width in rabbits' ileum across the treatments. Muscle thickness came out in similar trend observed in villus height, with the observed value for control lower (P<0.05) by 18.93%, 8.40% and 19.85% than rabbits fed probiotic, enzyme, and probiotic + enzyme supplemented diets respectively. The muscle thickness values obtained for the supplemented groups were similar (P>0.05). Just like in the duodenum, no significant difference existed in the ratio of villus height to crypt depth in the ileum.

Table 2: Intestinal morphology of growing rabbits offered corn cob-based diet supplemented with feed additives

Parameters	Control	Probiotic	Enzyme	Probiotic + Enzyme	SEM
Duodenum					
Villus height (µm)	411.80^{b}	516.03^{a}	494.17^{a}	503.96^{a}	15.73
Villus width (µm)	52.71	62.70	56.26	61.60	1.74
Crypt depth (µm)	90.38^{b}	114.44^{a}	108.12^{a}	116.36^{a}	3.58
Crypt width (µm)	42.05^{b}	51.21^{a}	48.02^{ab}	53.07^{a}	1.70
Muscle thickness (µm)	545.82^{b}	652.49^{a}	633.83^{a}	640.19^{a}	13.82
VH/CD	4.58	4.52	4.62	4.44	0.15
Ileum					
Villus height (µm)	470.03^{b}	705.03^{a}	622.09^{a}	667.47^{a}	28.89
Villus width (µm)	48.56^{b}	64.12^{a}	56.88^{ab}	62.21^{a}	2.46
Crypt depth (µm)	100.76	125.86	112.45	121.37	4.32
Crypt width (µm)	45.83	57.16	52.08	60.18	2.42
Muscle thickness (µm)	499.94^{b}	616.64^{a}	545.80^{a}	623.79^{a}	18.76
VH/CD	4.66	5.62	5.60	5.53	0.17

a, b, means in the same row with different superscript differ significantly (P<0.05). VH/CD = Villus Height/Crypt Depth

Discussion

The recorded morphological variations observed in the intestinal mucosa of duodenum and ileum via the significant increase in villus length, villus width, crypt depth, crypt width and muscle thickness in rabbits fed supplemented diets might be a means of meeting up with the increased rates of digestion brought about by activities of exogenous enzymes and probiotics. This equally might indicate that more surface area was needed for higher assimilation of nutrients provided in the gut of the treatments fed fortified diets. In the same vein, Buba and Shehu (2018) also submitted that greater villus height might lead to increase in the enzyme production from villi top with more enzymic activities bringing about improvement in digestibility. Therefore, the improved nutrients digestibility observed in the rabbits fed supplemented diets in this trial might not be unconnected to the improved gut morphology observed in them.

Under this study, villus height and muscle thickness in both duodenum and ileum of rabbits placed on supplemented diets were observed to be significantly higher than control. Crypt depth was equally found significantly higher in the duodenum of the rabbits offered treated diets in comparison to control, while the ratio of villus height to crypt depth was not significantly affected with the additives across the treatments. The results came up with in this work corroborated the report of Attia *et al*. (2012) when enzyme supplemented diet was fed to growing rabbits. Seyidoglu and Peker (2015) likewise observed significant increase in the villus height, depth of crypts, mucosa thickness with no significant difference in villus height ratio crypt depth of rabbits with increase in supplemented doses of yeast in the diets. Villus width obtained in this trial was found higher in the ileum of rabbits administered diets treated with probiotic and probiotic + enzyme than control. This result was similar to the report by Buba and Shehu (2018) who worked on baker's yeast supplementation in broiler chickens' diets and observed that villus perimeter and width were significantly improved in treatment fed diet supplemented with 2.0% *Saccharomyces cerevisiae* compared to control.

Conclusion and Application

It is therefore concluded that supplementing corn cob-based diet with probiotics (bactofort®) and exogenous enzyme (cellulase), either singly or combined improves intestinal morphology of growing rabbits which ultimately leads to improved utilization of nutrients in them.

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