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EVALUATION OF GROWTH PERFORMANCE OF RABBITS FED DIETS CONTAINING GRADED DIETARY INCLUSION OF GARLIC (*ALLIUM SATIVUM*) MEAL.

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ABSTRACT

Twenty-four grower rabbits were randomly assigned in a completely randomized design to four diets containing garlic meal at 0.00, 200, 400 and 600g/100kg diet to assess the effect of dietary inclusion of garlic on nutrient digestibility, caecal bacteria count, haematological and serum biochemical parameters. Data were collected on feed intake, faecal output, caecal bacterial count, haematological and serum biochemical indices of the rabbits. Result showed that crude protein and ether extract digestibility coefficients were higher ($p<0.05$) in groups fed diets containing 400g of garlic. Crude fibre digestibility coefficient was higher in rabbits fed diet containing 200g of garlic and least in rabbits fed control diet. *Bacillus* and *Lactobacillus* species were significantly higher ($p<0.05$) in rabbits fed diets containing garlic at 200g and 400g per 100kg diets followed by those fed diets containing 600g and least for control groups. *E. coli, Clostridium* and *Enterobacteriaceae* spp populations were significantly ($p<0.05$) higher in rabbits fed diet without garlic meal and lower in rabbits fed diets supplemented with garlic. Packed cell volume was highest ($p<0.05$) for control and least for groups fed diets containing garlic. Serum total protein (7.08 mg/dl), albumin (3.94 mg/dl) and globulin (3.15 mg/dl) were higher in rabbits fed diets without garlic supplementations. Glucose was higher in rabbits fed diets with garlic supplementations at 200g (195.68mg/dl), 400g (175.04mg/dl) and 600g (179.56mg/dl) per 100kg diets and least in the control groups (144.02 mg/dl). Total cholesterol contents were reduced in rabbits fed diets containing garlic at 600g (50.29mg/dl), 400g (59.88mg/dl) and 200g (74.10mg/dl) per 100kg diets compared with the control groups (91.13 mg/dl). conclusively, inclusion of garlic in the diets of growing rabbits up to 600g per 100kg improves nutrient utilization and beneficial caecal bacterial populations and had no detrimental effects on the animals.

Keywords: Digestibility; ceacal bacteria; haematological; serum profile; Rabbits

INTRODUCTION

The rising interest in the use of spices and herbs to supplement antibiotic growth promoters in livestock diets following ban by many countries of the World, has led to evaluation and use of bio-active botanicals some of these alternative feed supplements currently being promoted includes organic acids, prebiotics, probiotics, essential oils, and other medicinal plants products (Broz and Paulus, 2015). Phytochemical analyses of these botanicals have shown the presence of essential oil and phytochemicals that improve body function and overall performance (Hume, 2011).Garlic has been reported to enhance haematological parameters and nutrient use efficiency in rabbits (Onu and Aja, 2011). According to Sterling and Eagling (2001) supplementation of growing rabbit's diet with *Allium sativum* as phytobiotics reduces total blood plasma cholesterol, reduce blood pressure and decrease platelet aggregation .Enhancement of livestock diets with growth promoters is a common practice and according to Abdel-Wareth *et al*. (2014), it is used in rabbit diets to improve nutrient utilization. Among the numerous feed additives are phytobiotics such as garlic. It has been reported that garlic has antibiotic, antifungal, anti-parasitic, antiviral and antioxidant properties (Amagase *et al*., 2001; Tapiero *et al*., 2004; Shetty *et al*., 2013). "Herbs, spices, and plant extracts have been reported to enhance humans nutrition, making foods more appetizing and many of these herbs and spices are recognized for their health benefits. In addition to stimulation of appetite, they acts as antioxidants by mopping up free radicals and also supress microbial growth" (Al-Sultan, 2003; Al-Amin *et al.*, 2006; Al-Mashhadani, 2015)."These phytogenic plants could be used to replace some antibiotic growth promoters. To evaluate the efficacy of the phytobiotics as dietary supplements, they must be included to the feed in a more concentrated form than are found in their natural source" (Koch and Lawson, 1996). The study was therefore conducted to determine the effect of garlic supplementation as phytobiotics on nutrient digestibility, caecal bacterial, haematological and serum biochemical profile of growing rabbits.

MATERIALS AND METHODS

Study Location

The study was carried out at the Rabbit Unit of Samaru College of Agriculture, "Ahmadu Bello University, Zaria, Kaduna State. Zaria is in the Northern Guinea Savannah Ecological zone", located on longitude 11° 09' 01.78'' N and 7°39'14.79''E "671m above sea level. The climate is characterized by a well-defined dry and wet season. The climate is relatively dry with a mean annual rainfall of 700-1400mm (IARMS, 2019)".

Experimental design and rabbit's management

A total of 24 weaner rabbits (of about six to twelve months of age and 0.6kg to 1.0kg in weight) of mixed sexes and breeds were purchased from the National Animal Production Research Institute (NAPRI), Shika, Zaria, Kaduna State. They were fed on the experimental diets for a period of eight weeks. Clean water was supplied throughout the period of the study; other routine management practices were observed.

Experimental diets

The rabbits were randomly allotted to four dietary treatments replicated six times in a completely randomized design (CRD) as presented in Table1. Concentrate diets were supplied while 50g of chopped *Digitaria smutii* was supplied periodically to boost the dietary fibre level. Garlic meal was added as non-inclusive part of the diet as follows:

Diet 1: Standard diet without experimental material (Control)
Diet 2: Standard diet with garlic at200g/100kg
Diet 3: Standard diet with garlic at400g/100kg
Diet 4: Standard diet with garlic at600g/100kg

Table 1: Gross Composition of the basal diets fed to the growing rabbits

	Levels of *Allium sativum*(g/ 100 kg diet)			
Ingredients (kg)	**0**	**200**	**400**	**600**
Maize	35.00	35.00	35.00	35.00
Maize offal	27.00	27.00	27.00	27.00
Ground nut cake	15.00	15.00	15.00	15.00
Soya cake	20.00	20.00	20.00	20.00
Bone meal	2.00	2.00	2.00	2.00
Common salt	0.30	0.30	0.30	0.30
Methionine	0.25	0.25	0.25	0.25
Lysine	0.20	0.20	0.20	0.20
Vit-min premix	0.25	0.25	0.25	0.25
Total	**100.00**	**100.00**	**100.00**	**100.00**
Calculated analysis				
ME (kcal/kg)	2830	2830	2830	2830
Crude protein (%)	21.00	21.00	21.00	21.00
Crude fibre (%)	6.14	6.14	6.14	6.14
Ether extract (%)	3.46	3.46	3.46	3.46
Calcium (%)	0.46	0.46	0.46	0.46
Phosphorus (%)	0.80	0.80	0.80	0.80
Methionine (%)	0.41	0.41	0.41	0.41
Lysine (%)	0.71	0.71	0.71	0.71
Cost (₦/kg)	101.84	103.35	104.86	106.37

Vitamin premix supplied the following vitamins and trace elements per kg diet: vit A (7812.5 IU), vit D3 (1562.5 IU), vit E (25.0mg), vit K3 (1.25mg), vit. B1 (1.8mg), vit B2 (3.44mg), niacin (34.4mg), calcium pantothenate (7.19mg), vit B3 (3.1mg), vit B12 (0.02mg), choline chloride (312.5mg), folic acid (0.6mg), biotin (0.1mg), manganese (75mg), iron (62.5mg), zinc (50.0mg), copper (5.3mg), iodine (0.9mg), cobalt (0.2mg), selenium (0.1mg), antioxidant (75mg).

Digestibility studies

At the end of the study, two rabbits were randomly selected from each treatment for digestibility studies. Faecal samples were collected for five days and bulked; the bulked sampled were air dried and then were oven dried at 105^0C. The faeces collected for each treatment were analyzed for proximate composition: dry matter content (DM%), crude protein (CP%), ether extract (EE%), nitrogen free extract (NFE%), crude fibre (CF%) and ash (%) at the Biochemistry Laboratory of the Department of Animal Science, Faculty of Agriculture, Ahmadu Bello University, Zaria using the method described by A.O.A.C. (1990).

The percentage digestibility was calculated using the following equation:

$$\% \text{ Digestibility of Nutrients} = \frac{\text{Nutrient Intake} - \text{Nutrient Output}}{\text{Nutrient Intake}} \times 100.$$

Where, Nutrient Intake = % level of nutrient in feed × Feed consumed
Nutrient Output = % level of nutrient in faeces × Faeces output.
CP Output = (% CP in faeces × Faeces output) + CP in urine.

Caecal bacterial count

At the end of the study, the intestinal samples of two rabbits from each dietary treatment was taken after slaughtered. Samples of the caecal content were collected from approximately 3cm from the ileo-caecal junction. The samples were evaluated for bacterial species populations in the Microbiology Laboratory, Faculty of Veterinary Medicine, Ahmadu Bello University, Zaria.

Haematological studies

At the end of the study, 2mls of blood samples were taken respectively from each replicate into sterilised sample bottles with EDTA for haematological study. The samples were analyzed for differences in packed cell volume (PCV), haemoglobin (Hb) count, red blood count (RBC) and white blood count (WBC) as affected by the experimental material. The values recorded for PCV, RBC, WBC and Hb were determined using the Wintrobe'smicrohaematocrit, improved Neubauer haemocytometer and cyanometer-myoglobin method respectively.

Serum biochemistry

Another set of 2mls of blood samples were collected from each replicate into sterilised sample bottles without anticoagulant for serum biochemical profile and were assayed for total protein content, albumin, globulin, glucose and total cholesterol at the clinical pathology laboratory of Ahmadu Bello University Veterinary Teaching Hospital, Zaria.

Statistical analysis

Data collected were subjected to ANOVA. The difference between treatments means were separated using Duncan Multiple Range Test (SAS, 2002).

RESULTS AND DISCUSSION

Nutrient digestibility of grower rabbits fed diets supplemented with garlic (*Allium sativum)*.

The nutrient digestibility of grower rabbits fed diets supplemented with garlic as phytobiotics is shown in Table 2. There were significant differences ($p<0.05$) in all the parameters measured across the dietary treatments except for nitrogen free extract and dry matter digestibility coefficients which was not different ($p>0.05$).Crude protein and ether extract digestibility coefficients were significantly higher ($p<0.05$) in animals fed diets containing 400g of garlic. Crude fibre digestibility coefficient was higher in rabbits fed diet containing 200g of garlic and least in rabbits fed control diet. This implies that garlic inclusion in the diet of growing rabbits improves nutrient digestibility. This is in agreement with the findings of Gardzielewska *et al.,* (2003) who reported that supplementation of broiler chickens diet with garlic had strong stimulating effect on the immune system and also it's aromatic oils contents enhances digestion in birds.

Table 2:Nutrient digestibility of grower rabbits fed diets supplemented with garlic

	Inclusion Levels of garlic (g/100kg diet)				
Parameters	**0**	**200**	**400**	**600**	**SEM**
Dry matter (%)	55.63	55.30	55.15	55.96	0.76
Crude protein (%)	73.64^{c}	73.70^{c}	75.59^{a}	74.14^{b}	0.12
Crude fibre (%)	66.50^{d}	69.16^{a}	66.88^{c}	68.64^{b}	0.05
Ether extract (%)	62.15^{c}	65.44^{b}	66.67^{a}	56.27^{d}	0.21
NFE (%)	23.35	23.03	22.37	22.29	1.82

SEM: standard error of means, ^{abc}: means with different superscripts along the same row are significantly different ($p < 0.05$).

Caecal bacterial count of grower rabbits fed diets supplemented with garlic (*Allium sativum)*

Caecal bacterial count of growing rabbits fed dietary supplement of garlic as phytobiotics is shown in Table 3. There were significant differences ($p<0.05$) in the estimated bacterial populations except for *Salmonella* and *Pseudosomonas* spp. *Bacillus* and *Lactobacillus* species (gram-positive bacteria) were significantly higher in rabbits fed diets supplemented with garlic at 200g and 400g per 100kg diets followed by those fed diets containing 600g and least for control groups. *E. coli, Clostridium* and *Enterobacteriaceae* spp (gram negative bacteria) populations were higher in rabbits fed control diet and lower in rabbits fed diets supplemented with garlic. *E. coli,*

Clostridium and *Enterobacteriaceae* populations was least ($p<0.05$) in growing rabbits fed diet containing 600g of *Allium sativum* per 100kg diet. This implies that garlic (*Allium sativum)* supplementation in the diet of grower rabbits increases the population of beneficial bacteria and reduces the population of harmful microorganisms thereby improving the overall performance of the host organism. This observation is in consonance with the report of Lin *et al.* (2011) who reported that Phytonutrients, chiefly essential oil has been reported to "improve the intestinal micro biota of animals by enhancing the growth of beneficial bacteria species and suppressing the growth of potential pathogenic bacteria species". The result is in agreement with the findings of Lan *et al*., (2003) who reported that *Bacillius spp.* are one of the beneficial harmless microbes in the microbiota of livestock. The report by Aligiannis *et al*., (2001); Friedman *et al*., (2002) and Kirkpinar *et al*.(2011) are also in support of this study. These researchers observed "that essential oils such as oregano and garlic oil inhibit microorganisms like *E. coli* and other *Enterobacterial* counts, *Clostridium* count, *Staphylococcus aureus, Salmonella typhimurium* and *Listeria monocytogenes*)".

Table 3: Bacteria countof growing rabbits fed diets supplemented with garlic

	Levels of garlic (*Allium sativum)* (g/100kg diet)				
Bacteria ($\times10^3$cfu)	**0**	**200**	**400**	**600**	**SEM**
Bacillus spp.	3.80^{b}	5.65^{a}	5.65^{a}	4.15^{b}	0.24
Lactobacillus spp.	4.70^{c}	12.75^{a}	13.25^{a}	10.05^{b}	1.17
E. coli	0.93^{c}	0.05^{a}	0.37^{b}	0.00^{a}	0.12
Salmonella spp.	0.00	0.10	0.00	0.00	0.06
Clostridium spp.	14.20^{d}	9.05^{b}	11.15^{c}	0.45^{a}	0.76
Pseudomonas spp.	0.02	0.00	0.00	0.00	0.01
Enterobacteriaceae	1.15^{b}	1.15^{b}	0.00^{a}	0.00^{a}	0.02

SEM: standard error of means, ^{abc}: means with different superscripts along the same row are significantly different ($p < 0.05$).

Haematological profile of growing rabbits fed diets supplemented with garlic Haematological characteristics of growing rabbits fed diets containing garlic at different inclusion levels is shown in table 4. The result showed a significant differences ($p<0.05$) in most of the parameters measured except for white blood cells (WBC), lymphocytes, basophils, neutrophils and platelets which were not different ($p>0.05$). Packed cell volume (PCV) was significantly higher ($p<0.05$) in the animals fed on the control and least in the groups fed diets containing garlic. Red blood cells (RBC) and haemoglobin (Hb) were higher for the control groups. Most of the parameters measured were within the reference range for healthy rabbits except for lymphocytes which were above the normal range of 25-50g/dl (Mitruka and Rawnsley, 1997). It has been established that reduction in the levels of nutrients in feeds result in decrease in PCV and Hb of animals (Oladele and Ayo, 1999). "PCV is an index of toxicity, valuesl within the normal range" is an indication that the rabbits had the ability to tolerate the phytochemicals in the diet. The results obtained indicate that the diets contain adequate nutrients and hence supported their normal performance to maintain the normal haematological profile (Madubuike and Ekenyem, 2006). It also showed that the health of the rabbits was not negatively affected. Therefore, the diets would support oxygen transportation. This is in agreement with the findings of Roberts *et al.* (2000) who reported that diets of "poor quality would usually impair transportation of oxygen from the respiratory organs to the peripheral tissues".

Table 4: Haematology profile of growing rabbits fed diets supplemented with garlic

	Levels of garlic (*Allium sativum)* g/100kg diet)					Reference Value
Parameters re	**0.00**	**200**	**400**	**600**	**SEM**	
Packed cell volume	41.90[a]	35.85[b]	33.70[b]	35.00[b]	2.18	30.00-50.00
White blood cells	6.30	5.05	6.70	4.80	1.33	5.00-12.00
Lymphocytes	59.8	52.3	54.8	55.05	6.79	25.00-50.00
Eosinophils	3.30[b]	4.75[a]	4.25[ab]	3.95[ab]	0.62	0.00-5.00
Monocytes	9.65[b]	13.80[a]	10.75[b]	11.00[b]	0.67	-
Basophils	0.60	0.55	0.55	0.35	0.16	-
Neutrophils	26.65	28.6	27.95	29.65	2.76	35.00-55.00
Red blood cells	5.99[a]	5.45[ab]	4.75[c]	5.09[bc]	0.28	4.00-8.00
Haemoglobin	12.65[a]	11.20[b]	10.40[b]	10.95[b]	0.58	8.00-17.00
MCV	69.90[b]	65.80[d]	70.95[a]	68.65[c]	0.45	-
MCH	21.15[c]	20.60[d]	21.95[a]	21.55[b]	0.11	-
MCHC	30.25[c]	31.25[ab]	30.90[b]	31.40[a]	0.27	-
Platelets	340.00	290.00	136.00	234.00	81.65	-

SEM: standard error of means, [abcd]: means with different superscripts along the same row are significantly different ($p<0.05$), (Jenkins, 1993; Mitruka and Rawnsley, 1997; Ross *et al*., 1976).

Serum biochemistry of growing rabbits fed diets supplemented with garlic (*Allium sativum)*

Table 5 shows the serum indices of growing rabbits fed diets containing garlicat varying inclusion levels. Significant differences ($p<0.05$) were observed for all the parameters measured. Serum total protein (7.08 mg/dl), albumin (3.94 mg/dl) and globulin (3.15 mg/dl) were higher in rabbits fed diets without garlic supplementations.

Levels of glucose was higher in the treatment groups with garlic supplementations at 200g (195.68mg/dl), 400g (175.04mg/dl) and 600g (179.56mg/dl) per 100kg diets and least in the control groups (144.02 mg/dl). Total cholesterol contents were reduced in rabbits fed diets containing garlic at 600g (50.29mg/dl), 400g (59.88mg/dl) and 200g (74.10mg/dl) per 100kg diets compared with the control groups having the highest (91.13 mg/dl), indicating that with increase in the levels of garlic in rabbit diets, cholesterol content reduces. Sterling and Eagling (2001) also reported that garlic supplementation in livestock diets, lowers total plasma cholesterol and decreases platelet aggregation.

Table 5: Serum biochemical indices of growing rabbits fed diets supplemented with garlic

	Levels of garlic (*Allium sativum)* (g/100kg diet)				
Parameters	**0.00**	**200**	**400**	**600**	**SEM**
Total protein (mg/dL)	7.08[a]	5.77[b]	5.81[b]	6.12[b]	0.40
Albumin (mg/dL)	3.94[a]	3.03[b]	3.74[a]	3.53[ab]	0.26
Globulin (mg/dL)	3.15[a]	2.74[ab]	2.07[c]	2.59[b]	0.22
Glucose (mg/dL)	144.02[c]	195.68[a]	175.04[b]	179.56[b]	5.12
T. cholesterol (mg/dL)	91.13[a]	74.10[b]	59.88[c]	50.29[d]	2.92

SEM: standard error of means, [abc]: means with different superscripts along the same row are significantly different ($p < 0.05$).

CONCLUSION AND RECOMMENDATION

Within the scope of this study, it was concluded that garlic supplementation in diets of growing rabbits up to 600g per 100kg improves nutrient utilization and beneficial caecal bacterial populations while the population of the pathogenic bacteria decreased and has no detrimental effects on the general performance of the animals.

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