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Effects of ginger rhizome powder supplements and sex on hematological indices of prepuberal rabbits

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A 12 - week study was conducted to evaluate the effects of ginger rhizome powder (GRP) on hematological indices of prepuberal rabbits as well as to determine the effect of sex on their hematological indices. Seventy two rabbits of equal sexes were randomly divided into 4 groups (B_2, B_3, B_4 and B_1) of 18 (9 does and 9 bucks) each and fed diet with GRP at 5.0, 10, 15 and 0 g per kg feed in a randomized complete block design. All the hematological parameters were not significantly ($p>0.05$) affected by the diets. However, the packed cell volume and hemoglobin values of rabbits in B_2 and B_4 groups and the neutrophil counts of those on B_1 and B_4 groups were significantly ($p<0.05$) influenced by sexes. White blood cell count of the bucks on B_1 (12.80 $\times 10^6$ / µL) and B_4 (13.30 $\times 10^6$ / µL) diets were significantly ($p<0.05$) higher ($p<0.05$) than their doe counter parts. Lymphocyte values of bucks on B_1 (48.0%), B_3 (47.6%) and B_4 (57.2%) diets differed significantly ($p<0.05$) from their doe counterparts. The results suggest that supplementation of GRP up to 15 g per kilogram feed support blood cellular development in prepuberal rabbits.

Key words: rabbit, sex, ginger rhizome powder, hematology

Introduction

Over the years, antibiotics have been used as growth promoter in animal diets. It has been proven that its inclusion in the diet at low level is capable of enhancing growth performance in farm animals (Charis, 2000; Midilli *et al*., 2008). Its mode of action is to suppress the detrimental effects of pathogenic microbes in the gastrointestinal tract thereby reducing the occurrence of enteric diseases. However, the use antibiotics in controlling enteric diseases has been abused such that the residue of the antibiotics found their way into humans' food chain, hence, the European Union Commission placed a ban on the use of antibiotics as a growth promoter in animal diet (Cardozo *et al*., 2004; EUC, 2005).

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Due to the ban placed on the use of antibiotics, several other feed additive have been identified; these include; enzymes, microflora enhancer, immunomodulators, organic acids, prebiotics, probiotics, herbal products and combination of these products. The most prominent herbal products used in recent animal diet in Nigeria today include ginger.

Ginger is an underground rhizome of plant belonging to the family *Zingiberaceae.* Chemically ginger contains several classes of compounds. The chemical composition of dried ginger is as follows: starch 40-60%, proteins 10%, fats 10%, fibers 5%, inorganic material 6%, residual moisture 10%, and essential oil 1 - 4%. In all more than 200 different volatile substances have been characterized in the essential oil fraction wherein the pharmacologically active compounds are to be found. The volatile oil contains oleoresin, which is responsible for the pungency in ginger. The aromatic and pungent characteristics of ginger make it desirable in the culinary art. The economic importance of ginger centers on its use in the preparation of medicines.

The extracts of ginger have multiple pharmacological effects, some of which Guyer (2003) outlined to include inhibition of prostaglandin, thromboxane and leukotrienes synthesis. Ginger extracts are important in the inhibition of cold and, platelet aggregation. They are also important for their cardiotonic effects and gastro-intestinal actions. Ginger extracts have thermogenic and antibiotic activities and are important as digestive stimulants (Guyer, 2003). Ginger is used in food, beverages and confectionary (Rodriquez, 1971). It is also used industrially in the manufacture of items such as ginger ale, ginger beer, ginger wine, ginger bread, ginger essence, ginger chocolate pastries and biscuits.

Studies have shown that ginger can be used as a natural growth promoter as it enhanced immune function and favored meat quality in animals (Okoye *et al.*, 2006). Again, understanding the haematological constituents of rabbits fed diets supplemented with ginger rhizome powder is important, since such data indirectly reflect in the physiological responsiveness of the animals (Esonu *et al.*, 2001). It is against these backgrounds that this study was conducted to assess the hematological response of growing rabbits fed diets with graded levels of ginger rhizome powder supplement.

Materials and methods

Location: The experiment was carried out at the Rabbitry Unit of the Teaching and Research Farm, Department of Animal Science and Technology, Federal University of Technology, Owerri, located at latitude $4^0$4’ and $6^0$3' N and longitude $6^0$15' and 8^0 15'E. Owerri lies within the humid rainforest zone of west Africa characterized by long duration of rainfall and short period of dry

season with an annual mean temperature and relative humid of 27^0C and 80% respectively.

Experimental animals and management: Seventy two pre-puberal rabbits of mixed breeds (Chinchilla x New Zealand white) of equal sexes aged between 10 and 12 weeks were used for this study. The rabbit bucks weighed between 440.02 g and 445.74 g with average weight of 441.69 g, whereas the rabbit does weighed between 450.45 g and 450.62 g with average weight of 450.53 g. They were housed in individual cages measuring 1.5 m x 1.0 m x 1.0 m with wire mesh floor and wooden frames. They were fed commercial concentrate diet only for one week of acclimatization.

Processing of ginger rhizome powder: Fresh rhizomes of ginger of the Indian cultivar *Himachel Pradesh* were procured from National Root Crop Research Institute, Umudike, Abia State. The fresh ginger rhizomes were washed in water to remove adhering dirts. They were chopped into smaller pieces using kitchen knife. The chopped ginger rhizomes were dried under shade for 5 – 6 days to prevent volatilization of its essential oils by direct sunlight. The air dried samples were milled into fine particle sizes using Laboratory mill (Arthur Thomas, USA). The ground samples were sieved through 2 mm test sieve and there after incorporated into the diets.

Experimental diets: Four experimental diets were compounded with diet B_1 (control) containing no ginger rhizome powder while diets B_2, B_3, and B_4 contained 5 g, 10 g and 15 g ginger rhizome powder per kilogram feed, respectively. A randomized complete block design was used to assign the animals to the experimental diets such that there were 18 rabbits (9 does and 9 bucks) on each diet. The animals were fed the diets *ad libitum* with cool clean drinking water. The nutrient compositions of the experimental diets as shown in manufacturer's feed label are presented in table 1. The rabbits were fed the test diets for 12 weeks and at the end of the study, the animals were starved for 12 hours and thereafter killed by stunning and decapitation.

Blood collection and evaluation: At the end of the study, blood was sampled from the marginal ear vein of randomly selected animals for hematological analysis. They were collected into sterile vacutainer tubes containing a pinch of an anticoagulant, ethylene diamine tetra acetic acid for hematological analysis. Packed cell volume (PCV) and red blood cell (RBC) counts were determined as described by Ewuola and Egbunike (2008). White blood cell (WBC) and differential WBC count were determined as described by Schalm *et al.* (1975). Blood constants [mean corpuscular hemoglobin concentration (MCHC), mean corpuscular hemoglobin (MCH) and mean corpuscular volume (MCV)] were calculated using the appropriate formulae by Jain (1986).

Table 1. Analyzed chemical composition of the experimental diet

Nutrients	Percent (%)
Crude protein	18.00
Ether extract	6.00
Crude fibre	5.00
Salt	0.30
Calcium	1.00
Phosphorus	0.45
Lysine	0.75
Methionine	0.35
Metabolisable energy (Kcal / kg)	2900

Data analysis: Data collected were analyzed using the Statistical Analysis System (SAS, 1999) package using the procedures of Steel and Torrie (1980). Where significant differences were detected, means were compared using Duncan's New Multiple Range Test at 5% confidence interval. Statistical significance between sexes was determined by student's t - test.

Results and discussions

Hematological constituents reflect the physiological responsiveness of animal to its internal and external environment, which includes feed and feeding (Esonu *et al*., 2001). Table 2 showed the hematological indices of prepuberal rabbits fed diets supplemented with different levels of ginger rhizome powder. The RBC, PCV and Hb values were similar ($p>0.05$) among the treatment groups and also fall within the normal range of 3.7 - 8.0 10^6 µ / l, 25 – 50% and 8.9 – 17.5 g/dl reported for healthy rabbits by Mitruka and Rawnsley (1977). The normal PCV indicates the absence of normocytic anaemia which is reportedly characterized by normal MCV and MCH and only detected by a decreased number of RBC or PCV (Coles, 1986). The result is corroborated by the normal RBC which further elucidated the absence of hemolytic anaemia and depression of erythrogenesis. The normal hemoglobin concentration for all the experimental rabbits is probably an indication that ginger rhizome powder supplement supported hemoglobin synthesis, which according to Sirosis (1995) is among other factors, primarily affected by protein intake. The result suggests absence of microcytic hypochromic anaemia, which is due to iron deficiency and its improper utilization for the formation of haemoglobin.

Blood constant (MCHC, MCV and MCH) were similar ($p>0.05$) among the treatment groups but fall within the normal physiological range reported by Mitruka and Rawnsley (1977) for clinically healthy rabbits. This result suggests

that all the rabbits irrespective of the treatment had normocytic and normochromic red cell, meaning that the inclusion of ginger rhizome powder up to 15 g per kilogram feed did not affect iron utilization by these rabbits. However, the values returned for MCV and MCH in this study was lower than 86.60 fl and 29.57 pg reported by Ukorebi (2011) for MCV and MCH respectively in growing rabbits fed herb based diets. The lower MCH and MCV value could also be attributed to the breed used (Coles, 1986). The observed similarities in red blood cell indices partly agree with the report of Xianglu *et al*. (2009) who fed rats with ginger powder.

WBC and differential WBC counts are used as indicators of stress response and are sensitive biomarkers crucial to immune functions (Graczyk *et al*., 2003). WBC and differential WBC value were not significantly ($p>0.05$) affected by the dietary treatments among the groups. However, the WBC count fell within the normal physiological range reported by Mitruka and Rawnsley (1977). This indicates that body defense system of the rabbits in all the treatment groups was not negatively affected by ginger rhizome powder supplement. The increasing trend in WBC count of B_2 rabbits however, contradicted the findings of Xianglu *et al.* (2009), who reported a decreasing trend in WBC of rats administered ginger powder. High WBC count is usually associated with microbial infection or the presence of foreign body or antigen in the system (Ogbuewu *et al*., 2010b). The comparable mean WBC counts in all the treatment groups in this study ruled out the possibility of microbial infection. Also the comparable basophils, monocytes and eosinophil values with the reference values suggest that ginger rhizome powder supplementation do not improve the production of these blood parameters in rabbits.

The results of the effect of sex on hematological indices of pre-puberal rabbits fed diets supplemented with different levels of ginger rhizome powder additive are shown in table 3. Packed cell volume and hemoglobin values of the group on B_2 and B_4 diets were significantly ($p<0.05$) affected by sex. Even though there is a lack of literature on the effect of sex on hematological indices of prepuberal rabbits fed diets with varying levels of ginger rhizome powder with which our results could be compared, however, the PCV and Hb values of the males in $B_{1,}$ B_2 and B_3 treatment groups were significantly ($p<0.05$) higher than their female counterparts and these agree with the findings of Ogbuewu *et al*. (2010 a, b), Xianglu *et al*. (2009), Okeudo *et al.* (2003) and Kenneth and Carol (1998) in rabbits, wistar rats, local ducks and humans respectively. In addition, the significantly ($p<0.05$) higher PCV and Hb value of the B_4 female rabbits over their males agrees with the earlier findings of Oluyemi and Ologhobo (1998) and Nwosu (1979) in broiler birds but disagree with the findings of Ogbuewu *et al*. (2010 a,b), Xianglu *et al*. (2009), Okeudo *et al.*

(2003) and Kenneth and Carol (1998) in rabbits, wistar rats, local ducks and humans respectively. The PCV and Hemoglobin value in all the treatment groups fell within the normal range reported for healthy female and male rabbits by Milas *et al.* (2009).

Table 2. Effect of diets containing different levels of ginger rhizome powder supplements on hematological indices of prepuberal rabbits

Parameters	**Treatment**				**SEM**
	B_1	B_2	B_3	B_4	
RBC ($\times 10^{12}$ / L)	5.79	6.33	6.49	6.03	0.23ns
PCV (%)	36.53	35.98	36.83	36.03	0.71ns
Hemoglobin (g / dl)	12.65	12.58	12.85	12.53	0.28ns
MCHC (g / 100 mL)	34.60	34.90	34.83	34.40	0.22ns
MCH (pg)	21.93	19.93	19.88	21.23	0.61ns
MCV (fl)	63.55	57.10	57.05	61.58	1.93ns
WBC ($\times 10^9$ / L)	11.23	11.50	9.15	11.10	0.52ns
Lymphocytes (%)	54.28	61.00	56.23	53.43	2.26ns
Neutrophils (%)	44.83	39.00	46.03	46.58	2.22ns
Basophils (%)	0.00	0.00	0.00	0.00	0.00ns
Monocytes (%)	0.00	0.00	0.00	0.00	0.00ns
Eosinophils (%)	0.00	0.00	0.00	0.00	0.00ns

ns = significantly different at $p<0.05$; PCV- Packed cell volume; MCHC - Mean Corpuscular Hemoglobin Concentration; MCH – Mean Corpuscular Hemoglobin; MCV - Mean Corpuscular Volume; RBC- Red blood cell; WBC = White blood cell counts; B_1 = 0 g ginger rhizome powder per kilogram feed (control); B_2 = 5 g ginger rhizome powder per kilogram feed; B_3 = 10 g ginger rhizome powder per kilogram feed; B_4 = 15 g ginger rhizome powder per kilogram feed; SEM = Standard error of the mean.

Data on MCHC, MCH and MCV value were not significantly ($p>0.05$) affected by sex in all the treatment groups. However, female rabbits in B_1, B_3 and B_4 groups have relatively higher MCHC values over their males, whereas, males in B_2 treatment group maintained a reverse trend. The numerically lower MCHC value of females in B_2 group and MCH and MCV values of B_2 groups is at variance with the higher values of the same parameters obtained in females by Cetin *et al.* (2009) in rabbits. The comparable MCV, MCHC and MCH values between sexes in this study suggest that the values of red cell indices of prepuberal rabbits were not significantly ($p>0.05$) affected by sexes.

Males on B_4 treatment group had the highest WBC count which differed significantly ($p<0.05$) from their female counterpart. Neutrophil counts in B_1 and B_4 treatment groups and lymphocytes counts B_1, B_3 and B_4 groups were significantly ($p<0.05$) affected by sex. The observed higher WBC counts of males in B_1, B_2 and B_4 treatment groups relative to their female counterparts are in consonance with the findings of Ogbuewu *et al.* (2010a) and Bitto and

Gemade (2001) in rabbits but disagrees with the higher value of the same parameter reported in female by Okeudo *et al.* (2003). This disparity may be due to specie differences as reported by Okoro *et al*. (2011). The significantly higher lymphocyte counts of females in B_1 and B_3 groups in this study over their male counterpart was in accordance with the results of Cetin *et al.* (2009).

There is also no distinctive sex effect on the neutrophils in this study, but the neutrophil count of B_4 rabbits corroborates the result of Cetin *et al.* (2009).

Table 3. Effect of sex on hematological indices of pre - puberal rabbits fed diets containing different levels of ginger rhizome powder

Parameters		Treatment			
		B_1	B_2	B_3	B_4
RBC ($\times 10^6$ / L)	Male	5.46	7.00	6.62	5.60
	Female	6.13	5.67	6.35	6.45
	SEM	0.05	0.54	0.59	0.94
PCV (%)	Male	37.20	38.05[a]	37.25	32.30[b]
	Female	35.85	33.90[b]	36.40	39.75[a]
	SEM	0.73	1.47	1.45	2.27
Hemoglobin (g / dl)	Male	12.70	13.35[a]	13.00	11.00[b]
	Female	12.60	11.80[b]	12.70	14.05[a]
	SEM	0.05	0.54	0.59	0.94
MCV (fl)	Male	68.60	54.35	56.80	57.60
	Female	58.50	59.85	57.30	65.55
	SEM	4.17	1.67	1.68	6.41
MCH (pg)	Male	23.35	19.05	19.75	19.60
	Female	20.05	20.80	20.00	22.85
	SEM	1.07	0.61	0.48	2.18
MCHC (g / 100ml)	Male	34.10	35.05	34.80	34.00
	Female	35.10	34.75	34.85	34.80
	SEM	0.60	0.48	0.25	0.47
WBC ($\times 10^6$ / µL)	Male	12.80[a]	11.85	9.10	13.30[a]
	Female	9.65[b]	11.15	9.20	8.90[b]
	SEM	0.94	0.69	1.07	1.31
Neutrophils (%)	Male	52.00[a]	38.45	56.90	42.80[b]
	Female	37.65[b]	39.55	35.15	50.35[a]
	SEM	6.69	2.22	6.89	2.22
Lymphocytes (%)	Male	48.00[b]	61.55	47.60[a]	57.20[a]
	Female	60.55[a]	60.45	64.85[b]	49.65[b]
	SEM	5.80	2.22	6.84	2.22

[ab]Means within columns with different superscripts are significantly different ($p<0.05$).

Conclusion

The experiment showed that up to 15g ginger rhizome powder could be used as an additive in the diet of prepuberal rabbits without deleterious effects on blood cellular

components. This was based on the findings that ginger rhizome powder support growth and development of the blood cellular components in female and male prepuberal rabbits. Furthermore, it is recommended that a detailed research on pathophysiology of blood cellular components of rabbits fed *ad libitum* of the same supplemental levels to be conducted to ascertain its effects at micro-anatomy.

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