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Performance and Haematological Parameters of Rabbits fed graded levels of Bitter kola (*Garcinia kola*)

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The need to reduce cost of production of meat animal has necessitated the use of plant materials with medicinal properties as feed additives capable of minimizing the influence of pathogenic microbes and improving performance of the animal. This study investigated the effect of such medicinal plant materials *Garcinia kola* on the growth performance and the haematological parameters of 20 weeks old rabbits. Thirty six rabbits of mixed breed and mixed sexed were used for the experiment, which lasted for eight weeks (56 days). The rabbits were randomly assigned to four dietary treatments: T_1 with no (0%), T_2 (2.5%), T_3(5%) and T_4(7.5%) of *G. kola* inclusion. The rabbits were housed in 2m × 1m × 1m cages per three rabbits in a 4 × 3 CRD experimental design whereby each treatment had three replicates, with each replicate having 4 rabbits and each treatment comprised 12 rabbits. During the eight week period of the experiment, feed and water were given ad libtum and while similar managerial and sanitary measures were applied for all the animals. Daily feed intake and growth performance of animals in each unit were monitored. At the beginning and the end of the experiment, blood samples were collected from the ear and analyzed for haematological parameters Hb, PCV, RBC, WBC and Plasma proteins). The result showed that *G. kola* has significant effect on the feed intake, growth rate, haemoglobin, PCV, RBC, WBC and Plasma protein. The feed intake decreased in all rabbits on *G. kola* treated feeds; numerical values obtained were 332 ± 0.32, 285 ± 0.52, 288 ± 0.12 and 262 ± 0.33 g/week for treatment 1, 2, 3, 4 respectively. Weight depression was also observed similarly in all the rabbits on Garcinia treated feeds with T2 having weight loss of 0.36 ± 0.11, T30.35 ± 0.12 and T4 0.64 ± 0.10kg.
Rabbits on Garcinia treated feeds also have lower Hb, PCV, Plasma proteins and higher WBC and RBC compared to the initial values showing *G. kola*. *G. kola* appear to contain substances which depress feed intake and growth performance and the effect seem to increase with higher concentration. However, the RBC and WBC increased, it is therefore recommended that the use of *G. kola* should be at low levels of inclusion or intermittently not continuous.

Key words: *Garcinia kola,* growth performance, rabbits.

INTRODUCTION

Traditional animal healthcare practices involving use of some materials and herbal preparations called ethno-veterinary medicine is fast gaining grounds in the livestock industry especially in African and Asian countries (Ebenebe et al., 2010; Ojelade, 2015) as they provide readily available and low cost alternative to orthodox medicine. Of the herbal drug, the use of garlic (*Allium sativum*), ginger (*Zingiber officinale*), neem tree leaves

(*Azadiractha indica*) and most recently bitter kola (*Garcinia kola)* have been reported in livestock health care management (Ebenebe et al., 2010; Esonu 2006, Owen and Amakiri 2013 and Obun et al., 2013). *Garcinia kola* is a dicotyledonous belonging to the family Guttiferae or Clusiaceae and is widely cultivated throughout West Africa (Adedeji et al., 2006). In Nigeria, Otor et al. (2004) reported that *G. kola* is common in the South western states and Edo State. According to Chilaka (2009), *G. kola* is used for social, therapeutic and nutritional purposes. The pharmacological use of *G. kola* which is of utmost importance in many regions of Africa has been documented by many authors (Iwu et al*.,* 1993; Ofor et al., 2010; Okunji et al., 2007). Farombi et al. (2002) and Okunji et al. (2007) identified certain substances with antibacterial, anti-oxidative, anti-hepatoxic and hypoglycemic properties. Iwu et al. (1993) identified purgative, anti-parasitic and anti- microbial properties of *G. kola*.

The usefulness of *G. kola* in counteracting clinical and subclinical diseases that could hamper the performance of animals can only be ascertained by assessing its effect on growth performance of the animal and other haematological parameters. Owen and Amakiri (2015) noted that haematological parameters are indicators of the health status of animals and so constitute an indispensable tool in the diagnosis, treatment and prognosis of many diseases. Barnajee (2008) showed that haematological constituents of blood are valuable in monitoring feed toxicity especially feed constituents that affect the formation of blood. Ewuola and Egbunike (2008) also posited that haematological parameters appraise the health status of animals as they are indices of the effect of dietary treatments on the physiology of the animal. The most commonly used haematological parameters include Packed Cell Volume (PCV), White Blood Cell count (WBC), Red Blood Cell count (RBC), Haemoglobbin (Hb) and Plasma proteins (Dada and Ikeuerowo, 2009; Ahumibe and Braide, 2009). Mitruka and Rawnsby (1997) outlined normal ranges of haematological parameters for most animals. Esomonu et al. (2005) reported significant reduction in PCV, RBC and Hb in rats treated with 2 g/kg of *G. kola* in the 1st week of the trial but non-significant in the 2nd to 5th week while Dada and Ikeuerowo (2009) on the other hand reported non - significant change in the erythrocyte values of fish fed various concentrations of *G. Kola*. The result of aqueous, methanolic and ethanolic extract of *G. kola* in the haematological parameters of rabbits and rats has been fraught with inconsistencies. Osifo et al. (2013) investigated the effect of methanolic extract of *G. kola* on the haematological parameters of adult male rabbit and obtained significant reduction in the PCV, Hb, Neutrophil and eosinophil counts and significant increase in the WBC, Lymphocytes and Monocytes counts. Apart from haematological parameters, *G. kola* has been reported to depress feed intake and weight of rabbits (Uko et al., 2001; Notridge et al., 2008). However, the exact concentration that is detrimental to the performance and health of rabbits at the various stages of life is yet to be conventionally established. This study therefore investigates the effect of various concentrations of *G. kola* on growth performance and haematological parameters of 20 weeks old rabbits of mixed breed and mixed sexes fed diets containing graded levels of *G. kola.*

MATERIALS AND METHODS

Thirty six and twenty weeks old rabbits of mixed breeds and mixed sexes purchased from teaching and research farm of University of Nigeria, Nsukka (UNN) was transfered to the Rabbitry unit of the Department of Animal Science, Nnamdi Azikiwe University, Awka were used for the study. The rabbits were left in the cages to acclimatize for one week before being assigned to their respective dietary treatments. Before the on-set of the experiment, the rabbits were weighed individually and randomly assigned to labeled cages of 2 m × 1 m × 1 m in such a way that each cage housed male and female from each of the two breeds to counter the sex and age effect.

At the on-set of the experiment, blood was collected from the marginal vein of the ear in each of the rabbits, the blood was drained into blood vials containing Ethylene Diamine Tetra Acetic Acid (EDTA) bottles, while the blood for PCV was drained directly into labeled haematocrit capillary tubesto two-third full with one end of the capillary tube sealed with plasticine. The tubes were placed in microhaematocrit centrifuge and spun at 10,000 rpm for 5 min. Thereafter, the PCV was read with microhaematocrit reader and the readings were expressed in perecentage. For the WBC, the blood in the vial with EDTA was carefully drawn to 0.5 mark on white cell pipette and mixed thoroughly after the vial has been covered with finger tips. At an angle of 45°, the blood in the vial introduced into the improved Neubauer counting chamber (Haemocytometer) without allowing the fluid to overflow. The chamber was then placed on the microscope stage and allowed to settle for 10 min, so that using the 4mm objective and ×10 eye piece, all the cells were counted including cells touching the borderlines on the top and right hand side. For the RBC, the blood was also drawn to 5 mm mark on the pipette and made up to 101 mark (that is, to the point immediately above the bulb) with diluting fluid (3.0 g sodium citrate, 1.0 cm^3 formaldehyde and 100 cm^3 distilled water). Introduction of the blood into the counting chamber followed similar procedure as the case of WBC but counting was in five groups of 16 small sqaures in the centre millimeter square area that is 80 out of the 400 small sqaures. The haematological analysis was carried out at the Laboratory of the Zoology Department, Nnamdi Azikiwe University, Awka by Ufele A.N an animal physiologist in the research team. The rabbits were then subjected to dietary treatments containing 0% (T1), 2.5% (T2), 5% (T3) and 7.5% (T4) of *G. kola*, respectively.

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Table 1. Gross composition of the experimental diets.

Ingredients	Percent composition			
	T_1 (0%)	T_2 (2.5%)	T_3 (5%)	T_4 (7.5%)
Maize	40.0	39.00	38.0	38.0
Soybean Cake	7.00	6.00	5.00	4.00
Fish meal	1.00	1.00	1.00	1.00
Bitter Kola*	0.00	2.50	5.00	7.5
Wheat Offal	47.0	46.50	46.0	44.5
Blood Meal	1.00	1.00	1.00	1.00
Bone Meal	3.00	3.00	3.00	3.00
Lysine	0.25	0.25	0.25	0.25
Methionine	0.25	0.25	0.25	0.25
Salt	0.25	0.25	0.25	0.25
TM/Vit Premix	0.25	0.25	0.25	0.25
Total	**100**	**100**	**100**	**100**

Table 2. Proximate Composition of the Experimental Diet.

Parameter	T_1 (0%)	T_2 (2.5%)	T_3 (5%)	T_4 (7.5%)
Crude Protein (CP)%	16.38	16.06	15.73	15.34
Crude Fibre (CF) %	5.27	5.24	5.21	5.12
Ether Extracts (EE)	3.55	3.48	3.42	3.36
Ca %	1.25	1.29	1.33	1.37
P %	0.62	0.62	0.62	0.62
Lysine %	1.05	1.02	0.99	0.96
Methionine%	0.51	0.50	0.49	0.48
Energy (ME Kcal/Kg)	2486.50	2481.96	2477.42	2488.18

Other ingredients in the diet are shown in Table 1. The experiment was based on 4 × 3 CRD whereby each treatment had 12 rabbits, 4 rabbits per replicate and three replicates per treatment. The experiment lasted for 8 weeks (56 days) during which time the rabbits were given feed and water ad libtum and were subjected to similar sanitary and husbandry conditions. The daily feed intake was recorded while the weight records were taken on weekly basis using precision weighing balance for the eight weeks period of the experiment. At the end of the experiment, similar procedure described above was followed to collect blood samples for analysis of Hb, WBC, RBC and plasma proteins, as well as that for PCV analysis. Data generated from the experiment were analyzed statistically using ANOVA for CRD test while differences in means were assessed for significant differences using Duncan Multiple Range Test.

RESULTS

Proximate compositions of dietary treatment

The gross composition of the experimental diets is presented in Table 1 while the proximate composition is presented in Table 2. Table 2 presented percentage nutrient content of each of the dietary treatments. The dietary treatments are of similar protein and energy content such that the major source of variation is the inclusion level of *G. kola.* The crude protein content ranged from 15.34% in Treatment 4 with 7.5% level of *G. kola* inclusion to 16.38% in Treatment 1 with no *G. kola* inclusion. The energy content ranged from 24877.42 kcal/kg in Treatment 3 to 2488.18 kcal/kg in T3. However, there was no significant difference in the crude protein and energy content of the dietary treatments.

Proximate composition of *G. kola*

The result of the proximate composition of *G. kola* on Dry matter basis showed CP 2.64%, CF 20.51%, EE 9.47%, Ashes 1.07% and NFE 57.54%. The result showed that *G. kola* has very little protein, high fibre, ash and NFE.

Effect of dietary treatment on the performance of rabbits

The effect of the dietary treatment on the performance of the rabbits is presented in Table 3. The result showed

Table 3. Effect of the Dietary Treatment on the Performance of the Rabbits.

Parameter	T_1 (0%)	T_2 (2.5%)	T_3 (5%)	T_4 (7.5%)
Initial weight (kg)	1.58 ± 0.10	1.56 ± 0.15	1.53 ± 0.13	1.55 ± 1.00
Final weight (kg)	1.97 ± 0.12[a]	1.18 ± 0.14[b]1	1.08 ± 0.18[b]	0.91 ± 0.11[c]
Weight Gain/Loss (kg)	0. 39 ± 0.11	- 0.36 ± 0.11	-0.35 ± 0.12	- 0.64 ± 0.10
Feed Intake g/week	332 ± 0.32[a]	282 ± 0.52[b]	288 ± 0.12[b]	262 ± 0.33

+ S.E.

that the feed intake and weight was depressed progressively in all the rabbits subjected to *G. kola* based diet as the inclusion level increased from 2.5 to 7.5%. The final weight of the rabbits follows the opposite trend with the rabbits on control diet without *G. kola* treatment having highest final mean weight of 1.97 ± 0.12 kg and the least final mean weight of 0.91±0.11 kg in T4 with 7.5% thus, indicating weight loss on rabbits on *G. kola* treatment. Frutos (2004) reported that consumption of plant species such as *Garcinia kola* containing tannin (generally > 50g/kg^{-1} of DM) significantly reduced voluntary food intake in ruminants while medium or low consumption (> 50g/kg) seem not to affect it. Glick and Joslyn (1970) had earlier reported the food intake depression and subsequent decrease in weight of rats fed tannic acid. The decrease in food intake and weight loss in rabbits fed on diets with *G. kola* inclusion may therefore be associated with tannin content of *G. kola*. Although, Monago and Akhidue (2002) reported low content of tannins in *G. kola*, the percentage inclusion and period of feeding may be responsible for the result obtained in this study. *G. kola* also contain oxalate which is known to form a strong chelate with dietary calcium and other divalent metals and makes them unavailable for consumption (Abara et al., 2000). This coupled with inhibition of protein metabolism by tannins in *G. kola* may be responsible for weight depression in rabbits fed on *G. kola* in this study.

Haematological parameters of rabbits fed on *G. kola* based diet

The effect of dietary treatments on the haematological parameters of the rabbits fed on various levels of *G. kola* is presented in Table 4.

Effect of *G. kola* treatment on the haematological parameters

The result showed that the Hb values and plasma proteins of rabbits that received *G. kola* treatment were significantly ($P < 0.05$) lower than the values for the control. The Hb level for all treatments however was within the standard range 10.4 to 17.4 g/dl recommended for healthy rabbits (Mitruka and Rawnsley, 1977). Numerically, the Hb value for rabbits in the control was 11.50 ± 0.98 while that of treatment 2 to 4 were 10.02 ± 0.42, 9.50 ± 0.07 and 9.31 ± 1.06 g/dl constituting loss of 0.43, 1.15 and 1.00 g/dl in the respective *G. kola* treated feeds.

The result of plasma protein followed the same trend with loss of 0.15, 0.50 and 0.20 respectively. The PCV values obtained in the study were also within the normal range of 30 to 50% for healthy rabbits. There was significant difference ($P < 0.05$) in the PCV values obtained in all the treatments between the rabbits in the control and that of those in *Garcinia* treated feeds l with the highest loss in PCV value occurring in rabbits fed 7.5% *G. kola*. Esomonu et al. (2005) reported significant reduction in PCV, RBC and Hb in rats treated with 2 g/kg of *G. kola* in the first week of their trial but non- significant in the 2nd to 5th week of the trial. Osifo et al. (2013) also showed significant reduction ($P < 0.05$) in the PCV, Hb, neutrophil and eosinophil counts of rabbits fed with methanolic extract of *G. kola* and significant increase ($P < 0.05$) in the WBC and lymphocyte counts and an unchanged monocyte and basophil counts. The result therefore disagrees with Ahumibe and Braide (2009) who reported significant increase in PCV, Hb, and RBC in response to treatment with *G. kola*. Saponin content of *G. kola* may be responsible for these results. Monago and Akhidue (2002) remarkable high concentration of saponins in G. kola (15.79 ± 0.28 g/100 g DM). This concentration according to them may be deleterious when high concentration is consumed. Saponins induces haemolysis of erythrocytes (Onning et al., 1996), decrease in plasma cholesterol and bile acid production (Oakenful and Sidhu 1990). There is also an irreversible reaction of saponin with membranes of animals and cells as saponins render the cells non permeable. The dosage and saponin content of *G. kola* may be responsible for the inconsistent results from various authors. Besides, Monago and Akhidue (2002) reported high content of cyanogenic glycosides (59.56 + 0.05 mg/100 g DM) in *G. kola* which upon hydrolysis yields hydrogen cyanide (HCN) and thus, toxic at certain concentrations. HCN inhibit respiratory chain, therefore inhibiting metallo-enzymes such as cytochrome oxidases (Montgomery, 1980 cited in Monago and Akhidue, 2002). The study showed significant increase in WBC count in response to

Table 4. Mean Values of the Haematological Parameters from Each Dietary Treatment.

Haematological parameter		Treatment level inclusion			
		T_1 (0%)	T_2 (2.5%)	T_3 (5%)	T_4 (7.5%)
Hb g/dl	Initial	10.25 ± 0.35	10.45 ± 0.64	10.65 ± 0.71	10.35 ± 1.20
	Final	11.50 ± 0.98	10.02 ± 0.42	9.50 ± 0.07	9.35 ±1.06
	Gain/loss	1.25*	-0.43[b]	-1.15[a]	-1.00[a]
PCV (%)	Initial	33.50 ± 0.70	34.55 ± 0.70	34.60 ± 0.70	34.00 ± 2.83
	Final	33.80 ± 1.41	32.80 ± 2.12	32.75 ± 0.71	31.40 ± 0.71
	Gain/Loss	0.30	-1.75[b]	- 1.85[b]	- 2.60 [a]
WBC ($\times 10^5/mm^3$)	Initial	5.20 ± 0.28	5.20 ± 0.11	5.20 ± 0.28	5.20 ± 0.28
	Final	5.53 ± 0.57	5.90 ± 0.42	6.45 ± 0.49	7.40 ± 0.71
	Gain	0.33*	0.70[c]	1.25 [b]	2.20 [a]
RBC ($\times 10^6/mm^3$)	Initial	5.25 ± 0.35	5.15 ± 0.07	5.40 ± 0.21	5.45 ± 0.13
	Final	5.55 ± 0.77	5.35 ± 0.28	5.55 ± 0.31	5.30 ± 0.71
	Gain	1.30[a]	0.20[b]	0. 15[b]	0.15[b]
Plasma protein	Initial	42.50 ± 4.95	39.95 ± 4.95	41.85 ±1.95	41.80 ± 1.15
	Final	43.50 ± 2.12	39.80 ± 0.07	41.35 ± 0.35	41.60 ± 2.83
	Gain/Loss	1.00*	- 0.15[c]	-0.50[a]	-0.20[b]

Mean values are used, * Refer to gains Hb= Haemoglobbin, PCV= Packed Cell Volume, WBC= White Blood Cell, RBC= Red Blood Cell. The result showed decrease in the blood haemoglobin content and plasma protein of rabbits fed *G. kola*. The least record was in T3 with 5% level of inclusion, however, the WBC content increased progressively as the *G. kola* inclusion level in the diet increased.

G. kola treatment with the highest value recorded in Treatment 4 and the least in the control. The order of increase is T4>T3>T2>T1. The higher WBC recorded for the rabbit in the *G. kola* treated units could be attributed to the antimicrobial and anti-parasitic influence of *G. kola* effect of *G. kola* and the role it plays in the defense mechanism of the body of the animals. The result therefore suggests a well-adapted immune system for the treated groups. The result therefore corroborates the findings of Osifo et al. (2013) who reported significant reduction in PCV, Hb, neutrophil and eosinophil counts and significant increase in WBC and lymphocyte counts of rabbits fed on diets treated with methanolic extract of *G. kola.*

The result showed increase in RBC values for all rabbits in both the control and *Garcinia* treated rabbits but there was significant ($P < 0.05$) difference in the RBC values between the rabbits on the control diet and those on *G. kola* diets. The numerical values of the gain in RBC are 1.30, 0.20, 0.15 and $0.15 \times 10^6/mm^3$. Ahumibe and Braide (2009) reported increase in RBC for *Garcina* treated rats. Esomonu et al. (2005) who recorded increased RBC values in Wistar rats medicated with ethanol extract of *G. kola* seed. Unigwe and Nwakpu (2009) opined that the higher RBC counts for rabbits on bitter kola treatment could probably be due to compensatory action of the body whereby ageing RBCs were destroyed leading to release of some iron which in turn were salvaged and transported to the erythroid cells of the bone marrow for new haemoglobin and RBC syntheses.

Conclusion

The values of the haematological parameters obtained in this study were within the normal ranges thereafter, it appears that the effect of *G. kola* is not extremely detrimental to the life of the rabbit or the actions of any anti nutritional constituent of the seed, which does not exert long-term significant toxicological tendency to haematological parameters. However, *G. kola* use in rabbit production should be below 2.5% level of inclusion or be given intermittently to avoid its effect on feed intake and weight gain of the animals. *G. kola* effect on the body tissues especially the vital organs of the body should also be assessed in further studies. Besides, the findings of this study will also serve as a note of warning to many humans who are addicted to consumption of *G. kola* especially in this part of the world.

Conflict of interest

The authors have not declares any conflict of interest.

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