HAEMATOLOGY, SEROLOGY AND INTERNAL ORGAN CHARACTERISTICS OF RABBIT BUCKS FED *AMARANTHUS CAUDATUS* LEAF MEAL

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ABSTRACT

Some authors have proposed that Amaranthus caudatus leave can enhance the formation of the hemoglobin and improves blood profile. Furthermore, Amaranthus caudatus leave is high in energy, protein, carbohydrates, fat, vitamins, minerals and other trace elements. Thus, a Completely Randomized Design Experiment (CRD) was conducted to examine the blood profile, internal organs and carcass characteristics of New Zealand White rabbit bucks fed Amaranthus caudatus Leave Meal (ACLM). The treatments designated treatment 1 (T_1), treatment 2 (T_2) and treatment 3 (T_3) having 12 rabbits each were replicated 3 times with 4 rabbits per replicate. The age of the 36 prepubertal rabbit bucks was 3 to 4 months, and they weighed approximately 2.56 kg. Three diets formulated with ACLM and supplemented at 0, 10 and 20g/kg feed were fed to rabbits in the respective treatments. Data were collected for hematology, serology, internal organs and carcass characteristics of the rabbit bucks. Data collected on different parameters were subjected to analysis of variance (ANOVA). Results showed that significant increases (P<0.05) were observed on the Red blood cell (T_1 5.19; T_2 6.20; T_3 7.88 x10 ^{6}/mm^3), White blood cell (T_1 6.01; T_2 8.03; T_3 11.32 x10^9/mm^3), total protein (T_1 5.62; T_2 6.55; T_3 6.59g/dl), Glucose (T_1 69.06; T_2 71.20; T_3 73.90mg/dl) and Urea (T_1 22.15; T_2 25.77; T_3 25.83mmol/l). The carcass parts such as the Heart, Dressed percentage, Shoulder and Forearm significantly increased following the supplementation of ACLM. The serum total cholesterol significantly decreased as the level of supplementations increased. Thus, supplementation of ACLM at 10g/kg and 20g/kg in the diets of rabbit bucks improved some hematology and serum biochemistry parameters, internal organs and carcass characteristics of the New Zealand White rabbit bucks.

Keywords: *Haematology, Serology, Phytogenics, Internal organs, Red Amaranth, Rabbit.*

INTRODUCTION

The global interest in phytogenics has led to increased investigation of different plants than before. A systematic search for useful bioactivities from medicinal plants is now considered to be a rational approach in pharmaceutical and drug research. Plant materials, plant products and byproducts have continue to play important role in the maintenance of human and animal health since antiquity (Amaduruonye, *et al.,* 2017; Agida *et al.*, 2020). Currently, several plants and herbs are being used in part or whole to treat many diseases and to enhance animal nutrition and physiology (Ekuma, *et al.,* 2017; Uchewa, *et al.*, 2018; Jiwuba, *et al*., 2020). Active components, compounds and beneficial effects of these plant materials are now being in-

vestigated, extracted and developed into drugs or used as growth promoter in animal production (Oyeyemi and Ajani, 2014; Amaduruonye, *et al.,* 2018, Oguike *et al*., 2019). One of such plants is *Amaranthus caudatus.*

Amaranthus caudatus (Red Amaranth) is an annual or short-lived herbaceous perennial flowering plant, belonging to the family *Amaranthaceae* and of the genus *Amaranthus.* It is cultivated as a leaf vegetable, pseudo-cereals and ornament plant (Bensch, *et al,* 2003; Juan, *et al*, 2007; Ulbricht *et al.,* 2009; Peiretti, 2018). Red Amaranth is a rapidly growing herb, varying in height of up to 2 meters and above depending on the environment and rainfall in a cultivated lands and fallow in temperate and tropical climate. *A. caudatus* could be propagated through seed, stem and root stock (Svirskis 2003; Razaei *et al*., 2014; Peiretti, 2018). *Amaranthus caudatus* grows all year round with its peak in the rainy and early dry season in Nigeria. The red Amaranth produces purple, magenta or red flower and edible seed. Both the leaves, flower and seed are all edible to human and animal (Coastea, 2003; USDA, 2014). Red Amaranth is used as forage for farm animals, such as rabbits, sheep, goat and other livestock (Alegbejo, 2013; Seguin *et al*., 2013; Razaei *et al.,* 2013, 2014; Peiretti, 2018).

Some studies have shown that Red Amaranth leaves are high in carbohydrate, protein, fat and a good source of vitamins B, C, E, carotenoids and minerals such as- calcium, potassium, phosphorus, magnesium, manganese, iron, zinc and selenium (Martirosyan, *et al*., 2004; Babalola, *et al.,* 2010; Venskutonis, and Kraujalis, 2013; USDA, 2018). Some other authors have postulated that Red Amaranth has the ability to prevent fever, hemorrhage, and anemia and stomachache. It has been reported in several literatures to possess antioxidant, antimicrobial, anti-inflammatory, antitumoural, hypocholesterolemic and hemostatic properties in human (Svirskis, 2003; Ulbricht, *et al*., 2009; Alegbejo, 2013; Peiretti, 2018).

Red Amaranth is commonly used in many Nigerian communities as a vegetable. *A. caudatus* is a highly cherished edible vegetable plant and goes with most Nigerian starch dishes (O' Brien and Prince, 2008). Almost all tribes in Nigeria refers to the plant by differing names, such as Shoko or Shokoyokoto in Yoruba (meaning: make the husband fat), Alaiyoho in Hausa, Inine in Igbo (Alegbejo, 2013). In Isi-Ala Ngwa South, Abia State, Nigeria, red Amaranth is popularly used as blood supplement by boiling and the water extract consumed as tea.

Unlike its grain, Red Amaranth forage has received significant lesser research attention (Peiretti, 2018). As such, its overall potentials have not been fully elucidated. Therefore, it becomes justifiable to study the impact of Red Amaranth leaf meal on blood profile, internal organs and carcass characteristics of rabbit bucks and on its overall impact on the physiology of rabbit bucks.

MATERIALS AND METHODS

Experimental Location

This research was conducted in the Rabbitry Unit of the Teaching and Research Farm of the College of Animal Science and Animal Production, Michael Okpara University of Agriculture, Umudike, Abia State, Nigeria. Umudike is located in Abia state, Nigeria; at latitude $05^{0}29^{1}$ North and longitude $07^{0}31^{1}$ East; and at an altitude of 122 meters above sea level. It lies within the tropical rainforest zone of South Eastern Nigeria. The location is characterized by average annual rainfall of 2,177mm in 148- 155 rain days. The average ambient temperature is 25.5^{0}C with minimum and maximum temperature of 22^{0}C and 29^{0}C, respectively. Relative humidity ranged from 57-91% (NRCRI, 2010).

Collection and preparation of experimental diets

Red Amaranth leaves were collected from the demonstration farms of Michael Okpara University of Agriculture, Umudike, Abia State, Nigeria. It was air-dried under room temperature (26^{0}C) to a constant weight. The dry *A. caudatus* leaves were milled using a hammer mill to produce *A. caudatus* leaf meal. The *Amaranthus caudatus* leaf meal was used in supplementing the formulated diets. Three experimental rations were formulated containing 0g/kg, 10g/kg and 20g/kg *Amaranthus caudatus* leaf meal; Representing T_1, T_2 and T_3, respectively. The compositions of the experimental diets are presented in Table 1.

Table 1: Gross composition and calculated nutrients of experimental diet

Ingredients	Percentage Inclusion
Maize	44.94
Soya bean meal	17.31
Rice husk	32.00
Fishmeal	2.00
Bone meal	1.00
Limestone	2.00
Vit/min Premix*	0.25
Salt	0.50
Total	100.00
Calculated nutrients	
Crude Protein (%)	17.00
Metabolizable Energy (ME) (Kcal/kg diet)	2505.42
Crude fiber (%)	11.36
Lysine (%)	0.514
Methionine (%)	0.199

**Premix composition (per kg of diet): vitamin A, 12,500 IU; vitamin D3, 2500 IU; vitamin E, 50.00mg; vitamin K3, 2.50mg; vitamin B1, 3.00mg; vitamin B2, 6.00mg; vitamin B6, 6.00mg; niacin, 40mg; calcium pantothenate, 10mg; biotin, 0.08mg; vitamin B12, 0.25mg; folic acid, 1.00mg; chlorine chloride,300mg; manganese, 100mg; iron, 50mg; zinc, 45mg; copper, 2.00mg; iodine, 1.55mg; cobalt, 0.25mg; selenium, 0.10mg; antioxidant, 200mg.*

Experimental animals and management

Thirty-six (36) pre-pubertal New Zealand White rabbit bucks were sourced from the Teaching and Research farm of the College of Animal Science and Animal Production, Michael Okpara University of agriculture, Umudike, Abia State, Nigeria. A quarantine period of 2 weeks pre-experimental trial was allowed during which the animals were vaccinated against ecto and endo-parasite using Ivomectin and Levamisole at 0.1ml/kg body weight. The rabbits were randomly assigned to three experimental treatments and replicated three times with four rabbits per replicate in a completely randomized design. The experimental animals were housed in separate hutches throughout the experimental period. Feed and clean drinking water were provided *ad libitum*. Routine management practices were also carried out appropriately. The field work lasted for three months.

Experimental design

The design for the study was a Completely Randomized Design (CRD) trial with 3 treatments consisting of T_1, T_2 and T_3 respectively. T_1 served as the control. Twelve (12) rabbits were randomly assigned to each treatment, balanced for weights and replicated 3 times, with 4 rabbits per replicate. The respective experimental diets were *Amaranthus caudatus* leaf meal included at 0, 10 and 20g/kg feed. The experimental model is as follows:

$Y_{ij} = \mu + Ti + e_{ij}$

Where:

Y_{ij} = individual observation on the rabbit characteristics.
μ = overall mean
T_i = treatment effect
e_{ij} = random error assumed to be independently, identically and normally distributed with zero means and constant variances.

Data collection and evaluation

Weight determination

Initial and final body weights of the rabbits were measured on arrival at the experimental site. Subsequently, body weights and other growth performance parameters were taken at weekly interval throughout the study. All weight measurements were done using digital weighing scale (10kg capacity of 0.01 sensitivity). Weight gain was calculated as the difference between the final and the initial weight of the rabbits. At the end of the experiment, three rabbits sampled from each replicate (9 rabbits per treatment) were slaughtered using a captive bolt and allowed to bleed. The internal organs were collected, measured and recorded.

Haematology

Blood samples for hematological examination were collected from the ear vein of the rabbit bucks. A 5-ml syringe fitted with a sterile needle was used to collect about 2 ml of blood and quickly transferred to ethylenediaminetetraacetic acid (EDTA) sample bottles. The EDTA sample bottles were shaken gently to prevent clotting. The following hematological indices were determined: Packed cell volume, haemoglobin, Red Blood Cell, White Blood Cell, platelet, Mean Cell Volume, Mean Cell Hemoglobin and Mean Cell Hemoglobin Concentration. The platelets were determined according to the method as described by Bain *et*

al. (2017). Packed cell volume (PCV) was determined by the micro-hematocrit method as described by Kahn, *et al.* (2010). Hemoglobin (Hb) concentration was determined using a spectrophotometer through the Cyanomethanoglobin method as described by Putwain, (2008). Red blood cell (RBC) and white blood cell (WBC) counts were determined using Neubauerhemocytometer method as described by Bain *et al*. (2017). Mean corpuscular volume (MCV), mean corpuscular hemoglobin (MCH) and mean corpuscular hemoglobin concentration (MCHC) were calculated as described by Douglas and Wardrop, (2010).

Serum biochemistry

Two milliliters of blood was collected and transferred into sample bottles without anticoagulant. The blood was allowed to clot for 30 minutes, after which it was centrifuged at 3000 revolutions per minutes for 10 min in order to separate the serum from the clot. After the centrifugation, the serum was carefully collected and transferred into a clean sample bottle and the blood chemistry tests were performed thereafter. The following serum chemistry indices were determined as described by Kahn, *et al.,* (2010): Total protein (g/dl), bilirubin (mg/dl), creatinine (mg/dl); blood urea (mmol/l) and Alanine amino transferase (ALT) (µl). The blood for serum cholesterol and glucose were analyzed spectrophotometerically and colorimetrically, respectively as described by Stockbridge, *et al.* (1989) for cholesterol and Thomas and Labor (1992) for serum glucose. The blood samples for haematology and serum biochemistry were analyzed at the College of Veterinary Medicine, Michael Okpara University of Agriculture, Umudike.

Statistical analysis

Data collected on the different parameters were subjected to analysis of variance (ANOVA) in accordance with the methods of Steel and Torrie (1980). Significant means were separated according to Duncan's Multiple Range Test (Duncan, 1955).

RESULTS AND DISCUSSIONS

The results of the hematological analysis showed that the RBC and WBC were significantly ($P<0.05$) different compared to the control, while the other parameters were not. The Red blood cell and White blood cell increased on supplementation with *A. caudatus* leaf meal compared to the control. This suggests that the *Amaranthus caudatus* leaf meal improved the RBC and WBC. The red blood cells and white blood cells were still within the normal reference intervals for healthy rabbit bucks as recommended by Leineweber *et al.,* 2018. A closer examination

Table 2: Haematology of rabbit bucks fed diets supplemented with *Amaranthus caudatus* leaf meal treatments

Parameter	T_1	T_2	T_3	SEM
Blood pH	7.30	7.40	7.40	0.24
Red Blood Cell ($x10^6/mm^3$)	5.19^b	6.20^{ab}	7.88^a	0.31
Packed Cell Volume (%)	45.30	47.50	48.78	0.85
Hemoglobin (g/dl)	11.20	11.63	11.50	0.52
White Blood Cell ($x10^9/mm^3$)	6.01^b	8.03^b	11.32^a	0.89
Platelets ($x10^{9/}l$)	310.90	314.67	312.56	4.27
Mean Corpuscular Volume (fl)	77. 30	77.66	79.00	1.42
Mean Corpuscular Hemoglobin (pg)	20.10	20.76	21.00	1.33
Mean Corpuscular Hemoglobin Concentration (%)	25.48	25.56	26.19	2.84

^{abcd}: *means with different superscripts along rows are significantly different (P <0.05).*
SEM= Standard error of means

on the results also indicated that there were numerical improvements on most of the hematological parameters that were not significantly different.

The results of serum biochemistry of rabbit bucks fed diet supplemented with *Amaranthus caudatus* leaf meal are presented in Table 3.

Results on the serology showed that significant increases (P<0.05) were observed on the total protein, globulin, glucose and urea; while the serum total cholesterol decreases following the supplementation of *Amaranthus caudatus* leaf meal compared to the control. The reduction in serum total cholesterol is in line with the findings of Plate and Arêas, (2002), and Longato *et al*., (2017) who reported significant reductions on the cholesterol, triglyceride and serum lipid level in rabbits and broilers respectively, fed diets containing *A. caudatus* leaves compared with the control. These serum biochemical parameters were within the normal range as recommended by Putwain, (2008), Anna and Brigitte (2014). This inferred that the supplementation of *A. caudatus* leaf meal on the diets of the rabbits improved the serum biochemical parameters of the rabbit bucks. The results of the current study support the findings of Kabiri *et al*, (2010 and 2011) and Peiretti, (2018) who reported that *Amaranthus caudatus* leaves reduced the cholesterol level in hyper-cholesterolemic rabbits.

The results of relative organ weights of rabbit bucks fed diet supplemented with *Amaranthus caudatus* leaf meal are presented in Table 4.

The results showed that only the relative weight of the heart significantly increased (P<0.05) following the supplementation with *Amaranthus caudatus* leaf meal compared to the control.

Results of carcass characteristics of rabbit bucks fed diets supplemented with *Amaranthus caudatus* leaf meal are presented in Table 5.

From the results in Table 5, there were significant increases (p<0.05) on the dressed percentage, shoulder and forearm of the rabbit bucks fed

Table 3: Serum biochemistry of rabbit bucks fed diet supplemented with *Amaranthus caudatus* leaf meal treatments

Parameter	T_1	T_2	T_3	SEM
Total protein (g/dl)	5.62^{b}	6.55^{a}	6.59^{a}	0.14
Albumen (g/dl)	2.49	2.72	2.56	0.29
Globulin (g/dl)	3.14^{b}	3.82^{a}	4.02^{a}	0.20
Bilirubin (mg/dl)	1.61	1.69	1.81	0.32
Creatinine (mg/dl)	1.30	1.38	1.34	0.22
Glucose (mg/dl)	69.06^{b}	71.20^{b}	73.90^{a}	1.07
Total cholesterol (mg/dl)	106.34^{a}	95.97^{b}	90.05^{c}	1.13
Urea (mmol/l)	22.15^{b}	25.77^{a}	25.83^{a}	0.66
Alanine aminotransferase (µ/l)	18.29	18.57	18.94	0.84
Alkaline Phosphate (µ/l)	26.63	26.60	27.46	1.71
Aspartate aminotransferase (µ/l)	19.54	19.82	19.67	1.60
Calcium (mmol/l)	11.21	12.05	12.86	0.80
Phosphorus (mmol/l)	5.06	5.85	5.64	0.86
Potassium (mmol/l)	4.28	4.67	4.92	0.52
Sodium (mmol/l)	139.05	141.56	142.56	1.86

^{abcd}: means with different superscripts along rows are significantly different (P <0.05).
SEM= Standard error of means

diets supplemented with *Amaranthus caudatus* leaf meal compared to the control; while all the other parameters were not affected ($p>0.05$), although the supplemented groups increasing numerically with increased *A. caudatus* leaf meal inclusion. From these observations also, it could be inferred that *A. caudatus* leaf meal did not have adverse effect on the carcass parts of the rabbit bucks.

CONCLUSION

Based on the results and observations of this study, it is concluded that some hematological and serum biochemistry parameters, internal organs and carcass characteristics of New Zealand white rabbit bucks were significantly increased by supplementation of *Amaranthus caudatus* leaf meal especially at the higher levels of supplementation (ie T_3). It is therefore, recommended that Red *Amaranthus* leaf meal up to 20g/kg could be fed to growing New Zealand white rabbit bucks to improve blood profile, growth performance and carcass characteristics without deleterious effect on the physiology of rabbit bucks.

Table 4: Relative organ weights of rabbit bucks fed diet supplemented with *Amaranthus caudatus* leaf meal

Parameter	T_1	T_2	T_3	SEM
Liver (%)	2.12	2.17	2.34	0.03
Spleen (%)	0.02	0.02	0.03	0.02
Lungs (%)	0.22	0.24	0.27	0.01
Heart (%)	0.46^b	0.54^a	0.56^a	0.02
Right kidney (%)	0.26	0.25	0.25	0.02
Left kidney (%)	0.27	0.26	0.26	0.03

^{abcd}: means with different superscripts along rows are significantly different (P <0.05).
SEM= Standard error of means

Table 5: Carcass characteristics of rabbit bucks fed diet supplemented with *Amaranthus caudatus* leaf meal

Parameter	T_1	T_2	T_3	SEM
Live weight (g)	1934.54	1824.40	1821.39	38.59
Dressed weight (g)	1004.00	992.67	1000.00	16.76
Dressed percentage (%)	51.90^b	54.41^a	54.90^a	0.49
Drum stick (%)	3.08	3.30	3.26	0.07
Thigh (%)	10.06	10.51	10.45	0.14
Shoulder (%)	3.61^b	4.04^a	4.05^a	0.12
Forearm (%)	3.13^b	3.49^a	3.44^a	0.12
Breast part (%)	3.30	3.72	3.68	0.17
Back cut (%)	26.71	26.69	27.70	0.31

^{abcd}: means with different superscripts along rows are significantly different (P <0.05).
SEM= Standard error of means.

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