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Haematological and serum biochemical indices of rabbits fed different plant protein sources

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Abstract

The study was conducted to investigate the effects of feeding different plant protein sources on haematology and serum biochemistry of rabbits. Twenty (20) growing rabbits of mixed breed and sex with an average weight of 847.20g were randomly allotted to four dietary treatments with groundnut cake as treatment 1(T1), Moringa leaves as treatment 2(T2), Cowpea husk as treatment 3(T3) and groundnut haulm as treatment 4(T4) in a completely randomized design (CRD). At the end of the thirteen (13) weeks metabolism studies, blood samples were collected through the jugular vein of the rabbits and emptied into sample bottles containing ethylene diamine tetra-acetic acid (EDTA) and plain sample bottles for haematological and serum biochemical analysis respectively. The result on haematology showed significant ($P<0.05$) difference among treatment groups for haemoglobin (11.18 g/dL), white blood cell (5.10×10^9), neutrophil (28.33), eosinophil (1.33) and lymphocyte (70.00) with T2 (Moringa leaves) having higher values for WBC, eosinophil and lymphocyte. No significant ($P>0.05$) difference in red blood cell (RBC). There were significant ($P<0.05$) differences in Total protein, Aspartate transaminase (AST), Alanine transaminase (ALT), cholesterol, urea and creatinine. Total protein had a range of 59.67 g/l – 72.67 g/l with T1 (Groundnut cake) having significantly (($P<0.05$) higher value. Results obtained from this study showed that groundnut cake, moringa leaves, cowpea husk and groundnut haulm had no detrimental effect on haematological and serum biochemical indices. Therefore, they can be used in feeding rabbits.

Keywords: Blood, Groundnut, Haematology, Moringa and Rabbits

Introduction

High cost of conventional feedstuffs is one of the major limiting factor to large scale commercial rabbit production in Nigeria. In recent years, more people are involved in rabbit production but are faced with high cost of feeding. It has become imperative to develop appropriate and cost effective feeding systems for rabbit farmers. However, several authors have suggested that, the increasing scarcity of animal proteins and high cost of the conventional

feedstuffs in most developing countries can be addressed by incorporating plant feed such as forages and crop residues in the diets of rabbits (1).

Crop residues are the remains of crop plants that are left behind after the harvest. They are readily available sources of feed for livestock production. Nigeria as an agricultural country generates large tons of crop residues. Crop residues and by-products could result in environmental burden or pollution if not recycled or consumed by livestock. Efforts are being geared towards harnessing crop residues or by-products into animal feed (2 and 3). Rabbits are adaptable animals that can digest a wide range of plant materials, including crop residues. Some of the abundant crop residues are groundnut haulms and cowpea shells. The availability of these crop residues is always at peak immediately after harvest which falls between October-January of every year. Despite the demand for these crop residues for the ruminants, they still remain the cheapest and most available in the dry season. Reports on the use of groundnut haulms and cowpea shells in ruminants are well documented (1 and 5).

Growing rabbits require dietary nutrients for the growth and development of their body systems. The nutritional composition and digestibility of crop residues can be improved using different treatments and supplements to meet the requirements of growing rabbits (12).

Animal's blood parameters provide the opportunity to analyze its physiological, nutritional and pathological status and it aids in diagnosing nutritional and or environmental stress. As for the blood profile and serum biochemistry of rabbits fed on crop residues, it can vary depending on the specific crop residues used and the composition of their diet. Their blood profile would generally reflect their overall health and nutritional status, which can be influenced by factors such as the quality and balance of their diet, genetic factors, and environmental conditions (17).

Understanding the effects of these unconventional feed ingredients on the serum biochemistry and blood profile of growing rabbits is vital for assessing their potential as alternative protein sources and ensuring the overall health and productivity of the animals. Evaluating the serum biochemical parameters such as total protein, albumin, globulin, cholesterol, glucose, and urea levels can provide insights into the metabolic and nutritional status of the rabbits which are largely influence by the diets fed (18).

Many of these unconventional feed ingredients are fed to livestock without consideration to their health and physiological implication on the animal (11). A readily available and fast means of assessing clinical and nutritional status of the animal on feeding trial is the use of blood analysis (22). Haematological and serum profile are helpful in evaluation of rabbit's health and also provides valuable information on presence of metabolites and other constituents which are instrumental for detecting conditions of stress (4). This study is designed to evaluate the haematological and serum biochemical indices of rabbits fed different plant protein source.

Materials and Methods

Experimental site and location

The experiment was carried out at the Rabbit Unit of the Teaching and Research Farm of the Department of Animal Science, University of Maiduguri. Maiduguri is situated at latitude $11^0$5' North, longitude 30^0

0' East and an altitude of 354m above sea level. Maiduguri falls within the Sehelian Region (Semi-Arid Zone) of West Africa which is characterized by short duration of rainfall varying from 2-4 months (June-September). The rainfall varies from 300-500mm per annum and the ambient temperature rises from 25^0 C-28^0 C in December to January and higher in April and May which range from 39^0 C-40^0 C.

Source of experimental diets and materials

The feed ingredients used were groundnut cake, cowpea husk, groundnut haulm and *Moringa oleifera*. These ingredients were purchased dried from different open markets within Maiduguri metropolis and the rabbits were purchased from a small holder rabbit farmer within the Maiduguri metropolis. Maize, Maize offal, fish meal, plant protein source (groundnut cake, *Moringa*, cowpea husk and groundnut haulm), limestone, bone meal, vitamin premix, lysine and methionine were used to formulate a 13% crude protein diet. The plant protein source was used as the treatment with groundnut cake as treatment 1(T1), *Moringa* leaves as treatment 2(T2)**, Cowpea husk as treatment 3(T3) and groundnut haulm as treatment 4(T4) (Table 1).**

Management of experimental animals and design

Twenty (20) growing rabbits of mixed breed and sex with an average weight of 847.20g were randomly allotted to four dietary treatments (Groundnut cake meal as T1, *Moringa* leaves as T2, Cowpea husk as T3 and groundnut haulm as T4) with five rabbits per treatment each standing as a replicate. A completely randomized design was used in this experiment. The rabbits were housed in an individual cage which is raised 19cm above the ground, measuring 15cm×14cm×17cm (width× length ×height). The cages were provided with wire mesh floor which permit faeces and urine to drop. The rabbits were allowed one-week acclimatization period. The animals were provided with a feeder and drinker in each cage compartment. Each drinker was attached to the wire mesh to prevent wastage and wetting of the feed. The feeders, drinkers and the pen were cleaned daily. A total of 100g of formulated diet/animal/day was offered on dry matter basis. Daily left over feed was recorded and subtracted from the feed offered to determine the daily feed intake. Water was given ad-libitum. The rabbits were weighed to know their initial weight at the onset and weekly to the end of the feeding trial which lasted for 12 weeks.

Chemical analysis

The milled samples of groundnut cake, *moringa leaves,* cowpea husk and groundnut haulm and the experimental diets were taken to the central laboratory of the Department of Animal Science University of Maiduguri for proximate analysis. These samples were determined according to the procedure of (9). The following parameters were determined; dry matter (%DM), crude protein (%CP), crude fibre (%CF), ether extract (%EE) and ash (%Ash), and Nitrogen free extractive (%NFE). % NFE was calculated as;

% NFE = 100 - (% CP + % CF + % EE + % Ash).

Haematological and serum biochemical analysis

At the end of thirteen weeks of the metabolism studies, blood samples were collected through the jugular vein of the rabbit using heparinized or hypodermic

needle and syringe. Blood sample for haematological and serum biochemical analysis was emptied into sample bottles containing ethylene diamine tetra-acetic acid (EDTA) to prevent coagulation for haematological assay. While blood samples for serum biochemistry was collected into a plain sample bottle and left to coagulate for serum. The haematological indices determined were Packed Cell Volume (PCV), Haemoglobin (Hb), Red Blood Cell (RBC), White Blood Cell (WBC) and Differential Leukocyte (DLC). The biochemical parameters were direct bilirubin, conjugate bilirubin, total protein, globulin, Aspartate aminotransferase, Alanine aminotransferase, cholesterol urea and creatinine.

Statistical analysis

The data collected was subjected to analysis of variance (ANOVA), using the General Linear Model Procedure of (26). The significant differences among the treatment means was separated using (14) in the SAS Package.

Table 1: Ingredient composition of the experimental diets

Ingredients (kg/%)	Groundnut cake	*Moringa* leaves	Cowpea husk	Groundnut haulms
Maize	22	22	22	22
Maize offal	53	51	29	29
Fish meal	0	0	4	4
Limestone	1	1	1	1
Bone meal	2.5	2.5	2.5	2.5
Salt	0.5	0.5	0.5	0.5
Vitamin premix	0.5	0.5	0.5	0.5
L-Lysine	0.25	0.25	0.25	0.25
DI-Methionine	0.25	0.25	0.25	0.25
Wheat offal	10	10	10	10
Plant protein source	10	12	30	30
Total	**100**	**100**	**100**	**100**

RESULTS AND DISCUSSION

Proximate composition of experimental diets

The proximate composition of experimental diets presented in Table 2 showed the dry matter percentage of 93.1% in groundnut cake which is the highest value, while the lowest value was recorded in groundnut haulms 91.06%, the values obtained were lower than those reported by (18). The crude protein in groundnut cake was 13.99% which is the highest while cowpea husk had 12.13%. the CP in the test materials were adequate for maintenance as reported by (19). The crude fibre increased across the treatments from groundnut cake to groundnut haulms with the values of 10.36%, 10.69%, 14.86%, and 15.33% respectively. The values obtained were lower than those reported by (18) expect for cowpea husk and groundnut haulm which were similar. The variation observed might be due to the differences in the experimental diets which could be attributed to the varietal difference of the plant materials. The ether extract in *Moringa* leave meal was higher with the

Table 2: Proximate composition of experimental diets

Parameters (%)	Groundnut cake	Moringa leave	Cowpea husk	Groundnut haulms
Dry Matter	93.1	91.92	92.73	91.06
Crude Protein	13.99	13.55	12.13	12.72
Crude Fibre	1.94	2.8	2.08	1.84
Ether Extract	10.36	10.69	14.86	15.32
NFE	47.78	40.29	42.34	51.57
ME(Kcal/kg)	2370.96	2158.45	2120.36	2450.42

NFE = Nitrogen free extract, ME= Metabolizable energy

value of 2.8% and groundnut haulms having the lowest value of 1.84%. The nitrogen free extract of 51.57% was observed in groundnut haulms being the highest and the lowest in *Moringa* leave meal which was 40.29%.

Haematological indices

It is well established that changes in haematological parameters reflects the physiological status of the animal (16). The result of the haematological indices of rabbits fed different plant protein is presented in Table 3. The result showed significant ($P<0.05$) dietary effect on the haemoglobin (Hb) and white blood cell (WBC). The values for haemoglobin ranged from 11.18 to 13.20 which were higher than those obtained by (18) but within the normal range. Rabbits fed *Moringa* leave meal, had higher (5.10) WBC, cowpea husk and groundnut cake were statistically similar while groundnut haulm had the lowest value of 3.70. The range values of 3.70-5.10 obtained in this study falls within the normal range WBC ranges values of 2.5-12.5x 10^3 mm^3 for rabbits reported by (24). These results indicates that animals were healthy because if mean of WBC is below the normal range it is an indication of allergic condition, anaphylactic shock and certain parasitism, while elevated values (leucocytosis) indicates the existence of a recent infection usually bacteria. Packed cell volume (PCV) and red blood cell (RBC) means were not significantly influenced by the dietary treatments. However, the PCV (%) means for all the treatment groups fell within the normal physiological range of 30.0 to 50.0% as reported by (28).

The packed cell volume (PCV) is an index of toxicity reduction in the blood, and it suggests presence of a toxic factor which adversely has effect on blood formation. The PCV and RBC volume values (34-39%) and (5.93-6.13) obtained in this study fell within the reference range values of 30 to 50% for PCV and 4 to 7 for RBC respectively (28). This suggests that the diets were tolerated across the treatments. This agrees with the findings of (5), who reported that normal PCV and RBC value are indicators of adequate nutritional status of rabbits. This is an indication that the experimental diets fed were not detrimental to the health of the rabbits.

The white blood cell differential counts namely neutrophil, eosinophil, lymphocyte and monocytes were significantly ($p<0.05$) influenced by the dietary treatment. Rabbit fed groundnut cake had significantly ($p<0.05$) highest (42.00%) neutrophil count and lowest (22.00%) were observed in rabbit fed *Moringa* leave meal.

Rabbit fed *Moringa* leave meal had significantly ($p<0.05$) highest (74.00%) lymphocyte count and lowest (58.00%) were observed in rabbit fed groundnut cake. While basophil was not significantly ($p>0.05$) influenced by the dietary treatments. According to (15) the proportion of neutrophils and lymphocytes should be ratio 1:1, any deviation is an indication of infection. The range values of 28.33 to 38.33% and 61.33 to 70.00% recorded for neutrophils and lymphocytes in this study fell within 34 to 70% and 43 to 80% reported for rabbits (21). Haemoglobin functions in transporting oxygen to tissues of animal for oxidation of ingested feed so as to release energy for the other body functions as well as transport carbon dioxide out of the body of animal (27).

Table 3: Haematological indices of rabbits fed different plant protein

Parameters	Groundnut cake	Moringa leave	Cowpea husk	Groundnut haulm	SEM
Packed cell volume (%)	0.38	0.34	0.39	0.36	0.11^{NS}
Haemoglobin(g/dL)	12.53^{ab}	11.18^{c}	13.20^{a}	12.07^{b}	0.39*
White Blood Cells (x 10^9L)	4.73^{ab}	5.10^{a}	4.73^{ab}	3.70^{c}	0.20*
Red Blood Cells (x 10^{12}/L)	6.13	6.00	5.93	6.06	0.18^{NS}
Differential Leukocyte					
Neutrophil	37.60^{a}	28.33^{c}	38.33^{a}	32.67^{b}	1.33*
Eosinophil	0.00^{c}	1.33^{a}	0.33^{b}	0.67^{b}	0.25*
Lymphocyte	62.00^{c}	70.00^{a}	61.33^{c}	$66/67^{b}$	1.08
Monocyte	0.33^{a}	0.33^{a}	0.00^{b}	0.00^{b}	0.10
Basophil	0	0	0	0	0^{NS}

[abc]Means with the same superscript(s) within rows are not significantly different at 5% level of probability. SEM=Standard Error Mean

Serum biochemical Indices

Biochemical constituent reflects the physiological responses of the animal to its internal and external environments which include feed and feeding (17). The serum biochemical parameters of rabbits fed different plant protein sources showed significant ($P<0.05$) difference among the treatment groups for all the parameters. The direct bilirubin (DB) values of rabbits fed groundnut cake, cowpea husk and groundnut haulms(0.90, 0.93 and 1.03) were similar but, differed significantly ($P<0.05$) from the value (1.47) recorded for *Moringa* leave meal. The values were within the normal range of 0.86 to 5.16 mmol/L of direct bilirubin reported by (25). The conjugate bilirubin (CB) values of 1.83 and 1.67 mmol/L recorded in *Moringa* leave meal and cowpea husk were similar but differed significantly ($P>0.05$) from the values of 1.60 and 1.47 recorded in groundnut haulms and groundnut cake. Total protein (TP) values of groundnut cake is the highest (72.67 g/dL) followed by cowpea husk (68.67g/dL), but *Moringa* leave meal (59.67 g/dL) and groundnut haulms (59.67 g/dL) were similar. The Albumin values were similar in cowpea husk (36.00) and groundnut haulms (36.00 g/dL) but differed

significantly from groundnut cake (34.67 g/dL) and *Moringa* leave meal (31.33 g/dL). The range of values (31.33 to 36.00 g/dL) for albumin obtained in this study fell above the normal range reported of (2.5 to 4.5 g/dL) reported by (8). Abnormal serum albumin usually indicates an alteration of normal systemic protein utilization (10). Globulin is an important part of the immune system in rabbits, as it helps to fight off infection and disease. It is also involved in blood clotting and wound healing. Globulin also showed significant difference among the experimental groups with the highest significant value (38.00 g/dL) in groundnut cake. cowpea husk (32.67 g/dL), and groundnut haulms (32.00 g/dL) which are statistically similar but differed significantly from *Moringa* leave meal (28.33 g/dL). The values recorded in this study indicate that moringa leave meal is within the range (15 to 28 g/dL) while groundnut cake, cowpea husk and groundnut haulms are above the range as reported by (20). Both AST and ALT values obtained in this study are significantly ($P<0.05$) influenced by the experimental diets with groundnut cake having the highest value (33.00 U/L) and similar to *Moringa* leave meal but differed significantly ($P<0.05$) from cowpea husk (26.33 U/L) and groundnut haulms (17.67 U/L). Groundnut haulmsis having the lowest value (17.67 U/L) in AST, groundnut cake and cowpea husk are similar in ALT but significantly ($P<0.05$) differed from *Moringa* leave meal (55.67 U/L) and groundnut haulms (53.33 U/L). All the values obtained in this study for both AST and ALT fell within the normal range in rabbit AST (14-113 U/L) and ALT (48-80 U/L) (20). Both AST and ALT are enzymes that bare found in the liver, and they play an important role in the body metabolism. An elevated level of these enzymes is an indication of liver damage or dysfunction

The Cholesterol value in this study was significantly ($P<0.05$) affected by the treatment diets. There was trend towards a reduction in the cholesterol value across the experimental group. (1.83-1.27mmol/L) with groundnut cake having the highest value (1.83mmol/L) and significantly ($P<0.05$) differed from *Moringa* leave meal, (1.30 mmol/L) cowpea husk (1.27 mmol/L) and groundnut haulms (1.33 mmol/L). Cholesterol values obtained in this study falls within the normal range values (0.3-2.1mmol/l) (20). The urea values obtained in this study were significant ($P<0.05$) with groundnut haulmshaving the highest value of (6.13 mmol/L) and cowpea husk with the lowest value of (5.27 mmol/L) which is similar to *Moringa* leave meal (5.37 mmol/L) and *Moringa* leave meal is also similar with groundnut cake (5.67 mmol/L). All the values obtained in this study fell within the normal range (4.6-10.4 mmol/L) (20). Urea is a component of blood and a waste product that is produced by the breakdown of protein in the body, and it is filtered out in the blood by kidneys. High level of urea in rabbits indicates dehydration, kidney disease, urinary tract infections while low levels indicate malnutrition, and liver disease. Creatinine showed a significant ($P<0.05$) difference between the treatments. Groundnut cake recorded the highest value (109.67mmol/l) and the lowest value (68.67mmol/l) was recorded in cowpea husk. The values (68.67-109.67mmol/l) fell within the normal range for rabbits (44-221mmol/l (20). Creatinine is produced by the muscles, and it is filtered out of the blood by the

Table 4: Serum biochemical indices of rabbits fed different plant protein

Parameters	Groundnut Cake	Moringa leave	Cowpea husk	Groundnut haulm	SEM
Direct Bilirubin (mmol/L)	0.90[b]	1.47[a]	0.93[b]	1.03[b]	0.06
Conjugate Bilirubin (mmol/L)	1.47[b]	1.83[a]	1.67[ab]	1.60[b]	0.10
Total Protein (g/dL)	72.67[a]	59.67[c]	68.67[b]	59.67[c]	0.86
Albumin (g/dL)	34.67[b]	31.33[c]	36.00[a]	36.00[a]	0.63
Globulin (g/dL)	38.00[a]	28.33[c]	32.67[b]	17.67[c]	0.44
Aspartate transaminase (U/L)	33.00[a]	30.00[a]	26.33[b]	17.67[c]	2.01
Alanine transaminase (U/L)	62.00[a]	55.67[b]	62.33[a]	53.33[c]	1.11
Cholesterol (mmol/L)	1.83[a]	1.30[b]	1.27[b]	1.33[b]	0.09
Urea (mmol/L)	5.67[b]	5.37[bc]	5.27[c]	6.13[a]	0.17
Creatinine (mmol/L)	109.67[a]	98.00[b]	68.67[d]	86.00[c]	1.78

[abc]Means with the same superscript(s) within rows are not significantly different at 5% level of probability. SEM=Standard Error Mean

kidneys. An increase indicates kidney disease while decrease indicates malnutrition or severe muscle wasting.

Conclusion and Recommendation

Based on the result of this study it was concluded that groundnut cake, moringa, cowpea husk, and groundnut haulm had a positive impact on all the haematological parameters of rabbit except PCV. It is therefore recommended that rabbit farmers can use *moringa oleifera,* groundnut cake, cowpea husk, and groundnut haulm in rabbit farming.

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