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# HAEMATOLOGICAL AND SERUM BIOCHEMICAL INDICES OF WEANED RABBITS FED PAWPAW SEED OIL

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**Abstract:** This experiment was carried out to investigate the haematological and serum biochemical indices of weaned rabbits fed pawpaw seed oil. Thirty-six weaned cross bred rabbits (Chinchilla × New Zealand white) with an initial body weight of 517.0  $\pm$  0.26 grams were used for this experiment. Rabbits were balanced for weights and assigned to four treatment groups in a completely randomized design (CRD) in a feeding trial that lasted for eight weeks. Basal diet was formulated to meet the nutritional requirement of rabbits according to NRC (1977). Animals in treatment 1 was fed basal diet without pawpaw seed oil while treatment 2, 3 and 4 were fed same diet supplemented with pawpaw seed oil at 0.1 mL, 0.2 mL and 0.3 mL per kilogram respectively. Feed and water were supplied at all times. Results obtained showed that pack cell volume, haemoglobin, red blood cell, mean corpuscular volume, mean corpuscular haemoglobin, mean corpuscular haemoglobin concentration, white blood cell, lymphocytes, monocytes and basophil count which varied from 27.82 – 34.65 %, 8.45 – 13.06 g/dL, 4.81 – 6.07 (×10<sup>6</sup>μL), 38.83 – 54.11 fl, 16.54 – 26.57 pg, 60.46 – 82.60 %, 7.45 – 11.56 (×10<sup>3</sup>μL), 54.73 – 70.67 (×10<sup>3</sup>μL), 5.17 – 5.30 (×10<sup>3</sup>μL) and 2.13 – 3.10 (×10<sup>3</sup>μL) were influenced (P<0.05) by the treatment. Albumin, globulin and glucose levels were significantly (P<0.05) increased except for creatinine and urea values (P>0.05). However, values obtained were within the established range for healthy rabbits. In conclusion, pawpaw seed oil can be fed to weaned rabbits up to 0.3 mL per kg without causing any deleterious effect on their health status.

Keywords: Pawpaw; Oil; Rabbits; Haematology; Serum

# 1 INTRODUCTION

Investing and promoting rabbit production is one of the ways to meet up with the increasing human demands for quality protein, since beef, Pork and Chicken have insufficiently met human protein requirement. It has been observed that recently, there has been increased awareness in rabbit production because of their high prolificacy, rapid maturity, high genetic potential, efficient feed utilization, limited competition with humans for food and high-quality nutritious meat [1]. Rabbits have been notably introduced into West Africa as farm animals of economic value, with its high quality meat low in fat, succulent and providing a palatable change for chicken and other meats, [2]. The qualities possess by rabbit meat make it suitable animal protein for people living with cardiovascular diseases who are on low fat and sodium diet [3]. More so, rabbit meat has no religious taboo regulating its consumption unlike some animals whose consumption is restricted due to religious and ethical beliefs. These therefore, make rabbit a good source to bridge the gap of inadequate intake of animal protein [4]. Varieties of herbs and ethnobotanical plants parts have been explored to boost productivity in many animal species. This is due to their versatility and richness in various bioactive compounds which helps to maintain good health.

Papaya belongs to family Caricaceae and it is a native of tropical America but now spread all over the tropical world [5,6]. Traditionally, the leaves, seeds, latex and fruit have been used for the treatment of malaria, dengue, jaundice, warts, sinuses, eczema, cutaneous tubercles, blood pressure, dyspepsia, constipation, amenorrhoea and expel worms [7,8]. The plant is a good source of vitamin A, B and C and it is also loaded with several bioactive phyto-constituents including alkaloids, glycosides, flavanoids, saponins, tannins, phenols, steroids, amongst others [7,9]. This phyto-constituents enable the plant to perform multiple biological roles such as, anti-inflammatory, hypoglycaemic, anti-fertility, hepato-protective, wound healing, antihypertensive and antitumor activities [7].

Isaac *et al.*; Iheukwumere and Herbert reported incidences of toxicity among animals fed diets containing phytogenic compounds, hence, the need to investigate nutritional composition[10,11], phytogenic properties and the applications of these plant materials before recommending their use in animal production. The clinical examination of blood becomes necessary since it provides reliable information about feed toxicity on animals fed such diets and an assessment of the animal's growth also becomes imperative.

Therefore, this experiment was designed to determine the haematological and serum biochemical indices of weaned rabbits fed pawpaw seed oil.

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#### 2 MATERIALS AND METHODS

## 2.1 Experimental location

This trial was carried out at the Animal Science Unit of the Teaching and Research Farm, University of Abuja, Abuja, Nigeria. The site lies between latitude 80 56'29" North, 70 5' 31" East, and longitude 0070 20' North and 0070 51' East. The experiment was carried out according to the specifications and guidelines of animal and animal protocol approved by the Research and Ethics Committee of the Department of Animal Science, University of Abuja, Nigeria.

## 2.2 Collection and processing of pawpaw seed oil

Ripe pawpaw fruits were harvested from University of Abuja, Teaching and Research Farm and taken to the Department of Biological Sciences for identification and authentication by a certified taxonomist and assigned a voucher number (AD/021/007). Seeds were collected from the ripe fruits, air dried for 10 days and milled using an electronic blender. 200 grams of grinded sample was placed in a sealed filter paper and set in a Soxhlet apparatus for the extraction of oil. Collected oil sample was sent to the laboratory for further examination.

### 2.3 Animal care and experimental design

A total of thirty-six weaned rabbits (Chinchilla  $\times$  Newzealand white) with an initial body weight of  $517.0 \pm 0.26$  grams were used for this experiment. The rabbits purchased from a reputable source, placed on two weeks' quarantine period, fed basal diet which was adequate in all nutrients according to Nutritional Research Council's recommendation in 2007 and also treated against parasites using Albendazole® (1 tablet to 500 grams' body weight). The experimental animals were housed individually in a wired hutches measuring 100 cm by 80 cm by 40 cm (length  $\times$  width  $\times$  height) raised above the floor. All experimental animals were subjected to the same housing and management conditions. The rabbits were balanced for weights and assigned to four treatment groups in a completely randomized design (CRD) in a feeding trial that lasted for (8) weeks. Rabbits in treatment 1 was fed basal diet without pawpaw seed oil while treatment 2, 3 and 4 were fed same diet supplemented with pawpaw seed oil at 0.1 mL, 0.2 mL and 0.3 mL per kilogram respectively.

# 3 Proximate evaluation of experimental diet

Near infra-red kit (Model: TNOP NIRS<sup>TM</sup>, Netherlands) was used to analyze experimental diet. 300 grams of sample was placed in a collection tray after putting on the start button and kit was operated according to the manufacturer's recommendation. Result was obtained within 60 seconds and printed out from the visual display unit. To ensure precision in results, optical band, data resolution and wavelength was maintained at 9.00 nm, 0.6 nm and 2600 nm respectively.

# 3.1 Blood analysis

At the end of the trial, 4 mL of blood samples was collected from the ear marginal vein of five randomly selected rabbits per treatment. 2 mL of blood was transferred into a labeled sample bottle with anticoagulant (ethylene diamine tetra acetic acid) and placed in an ice pack before it was to the laboratory for further analysis. Parameters examined include, pack cell volume, red blood cell, haemoglobin, white blood cell, basophils, monocytes and lymphocytes. Samples were analyzed using Systmex automatic haeamtology analyzer (Model NH/118f, India) and maintained at an ambient temperature of  $18 - 32^{\circ}$ C and humidity of 80 % to ensure accuracy in result. The other 2 mL blood sample was put in a sample bottle without anticoagulant for serum biochemical analysis. Examination was carried out using Aura blood commercial analyzer (model HH/0u/2021A, China). Calibration was done according to the manufacturer's recommendation to ensure precision in results.

# 3.2 Data analysis

All data collected were subjected to one-way analysis of variance (ANOVA) using SAS and the significant means separated by Duncan's multiple range test at 5 % level of significance[12].

#### 4 RESULTS AND DISCUSSION

**Table 1** Ingredients and Chemical Composition of Experimental Diets

Materials	Diet 1	Diet 2	Diet 3	Diet 4
Maize	55.00	55.00	55.00	55.00
Wheat bran	10.40	10.40	10.40	10.40
Soybean meal	25.05	25.05	25.05	25.05

Fish meal	1.00	1.00	1.00	1.00
Limestone	2.05	2.05	2.05	2.05
Bone meal	4.00	4.00	4.00	4.00
Lysine	0.20	0.20	0.20	0.20
Methionine	0.30	0.30	0.30	0.30
Vitamin/Mineral premix*	0.25	0.25	0.25	0.25
Salt	0.30	0.30	0.30	0.30
Pawpaw seed oil (mL/kg diet)	0.00	0.10	0.20	0.30
Total	100.00	100.00	100.00	100.00
Determined analysis (%)				
Crude protein	17.06	17.06	17.06	17.06
Crude fibre	10.92	10.92	10.92	10.92
Ether extract	3.14	3.14	3.14	3.14
Calcium	1.15	1.15	1.15	1.15
Phosphorus	0.73	0.73	0.73	0.73
Metabolizable energy (Kj/kg)	10.09	10.09	10.09	10.09

Vitamin-Mineral Premix: (Rotinol) based on 2.5 kg/ton (Thiamine, 1500 mg, riboflavin, 5000 mg, pyridoxine, 5000 mg, cyanocobalamine, 25 mg, niacin, 60,000 mg, D-panthotenate, 20,000 mg, folic acid, 200 mg, D-biotin, 8 mg, Retinyl acetate, 40 mg, cholecalciferol, 500mg, tocopherol acetate, 40,000 mg, menadione, 800 mg, ascorbic acid, 60,000 mg, manganese, nill, iron, 80,000 mg, zinc, nill, copper, nill, cobalt, 80 mg, iodine, 400 mg, selenium, 40 mg, choline chloride, 80,000 mg

Effect of pawpaw seed oil on the haematological parameters of weaned rabbits is presented in Table 2. The dietary treatment influenced (P<0.05) the packed cell volume, haemoglobin, red blood cell, mean corpuscular haemoglobin, mean corpuscular volume, mean corpuscular haemoglobin concentration, white blood cell, monocytes, lymphocytes, basophils and neutrophils. Packed cell volume, haemoglobin, red blood cell count took the form of 27.82 - 34.65 %, 8.45 - 13.06 g/dL and 4.81 – 6.07 (×10<sup>6</sup>μL) respectively. The obtained values were within the normal range (22.00 – 36.00 %), 6.50 – 15.00 g/dL and 4.00 – 8.00 (×10<sup>6</sup>μL) cited by John; Jain [13,14]. This suggests efficient supply of oxygen in the tissues which gives room for better nutrient utilization in the body of rabbits [15]. Low pack cell volume, red blood cell and heamoglobin count is an indication of aneamia. Mean corpuscular haemoglobin, mean corpuscular volume, mean corpuscular haemoglobin concentration varied from 16.54 – 26.57 pg, 38.83 – 54.11 fl and 60.46 – 82.60 % and were within the normal ranges reported by Jain [16]. The result obtained is in consonance with the reports of Oloruntola et al. when Mucuna pruriens leaf meal is supplemented in the diets of growing rabbits[17]. Mean corpuscular haemoglobin and mean corpuscular haemoglobin concentration decrease during cases anaemia or low iron concentrations in the blood [18,19]. White blood cell, monocytes, lymphocytes, basophils and neutrophils count took the form of  $7.45 - 11.56 \,(\times 10^3 \mu L)$ , 5.17 - $5.30 (\times 10^3 \mu L)$ ,  $54.73 - 70.67 (\times 10^3 \mu L)$  and  $2.13 - 3.10 (\times 10^3 \mu L)$  were within the established range reported by John (2024); Brown and Clime [20,21]. The presence of phyto-constituents in pawpaw seed oil could trigger the production of antibodies making rabbits less susceptible to disease [22,23]. Monocytes and lymphocytes are capable of strengthening the immune system [22,24]. Basophils counts are triggered by parasites or allergic conditions [23].

Table 2 Effect of Pawpaw Seed Oil on the Haematological Parameters of Weaned Rabbits

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Variables	T1	T2	Т3	T4	SEM
Pack cell volume (%)	27.82°	32.74 <sup>b</sup>	33.48 <sup>b</sup>	34.65ª	0.60
Haemoglobin (g/dL)	8.45°	11.58 <sup>b</sup>	12.48 <sup>a</sup>	13.06ª	0.42
Red blood cell (×10 <sup>6</sup> μL)	4.81 <sup>b</sup>	5.55ª	5.56ª	6.07 <sup>a</sup>	0.13
Mean corpuscular volume (fl)	38.83°	44.95 <sup>b</sup>	52.42ª	54.11ª	1.54
Mean corpuscular haemoglobin (pg)	16.54°	22.48 <sup>b</sup>	23.79 <sup>b</sup>	26.57ª	0.90

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Mean corpuscular haemoglobin concentration	60.46 <sup>b</sup>	64.62 <sup>b</sup>	80.60a	82.60ª	4.84
(%)					
White blood cell (×10 <sup>3</sup> μL)	7.45 <sup>b</sup>	10.60 <sup>a</sup>	11.48ª	11.56ª	0.40
Lymphocytes (×10 <sup>3</sup> μL)	54.73 <sup>d</sup>	56.41°	68.57 <sup>b</sup>	70.67ª	3.39
Monocytes (×10 <sup>3</sup> μL)	5.30°	5.75 <sup>a</sup>	5.32 <sup>b</sup>	5.17 <sup>d</sup>	0.07
Basophils (×10³μL)	2.13 <sup>b</sup>	2.67 <sup>ab</sup>	2.90ª	3.10 <sup>a</sup>	0.11

Means in the same row with different superscripts differ significantly (P<0.05); T<sub>1</sub>: basal diet without pawpaw seed oil; T<sub>2</sub>: Basal diet with 0.1 mL pawpaw seed oil; T<sub>3</sub>: Basal diet with 0.2 mL pawpaw seed oil; T<sub>4</sub>: Basal diet with 0.3 mL pawpaw seed oil; SEM: standard error of mean

Table 2 reveals the effect of pawpaw seed oil on the serum biochemical indices of weaned rabbits. The dietary treatment influenced (P<0.05) albumin, globulin and glucose except for creatinine and urea levels. Albumin and globulin values were higher in treatment 3, 4 and lower in treatment 1 and 2 (P<0.05). Values recorded for albumin and globulin in this experiment were within the normal range 1.90-4.50 g/dL and 1.30-3.00 g/dL cited by Mitruka and Rawnsley[25]; John phytogenics were fed to weaned rabbits[26]. Oloruntola et al. recorded a higher albumin value of 2.00-4.50 g/dL in rabbits fed diet supplemented with Gliricidia leaf meal[17,27]. This variation could be attributed to the composition of phytoconstituents in test ingredients [28]. It may also suggest adequate protein intake and uncompromised health status of rabbits. Creatinine, urea and glucose values were within the normal range 45.00-81.00 (µmol/L), 3.00-10.00 Mmol/L and 5.00-8.00 Mmol/L cited by Kaneko[29]. Creatinine and urea levels recorded in this study suggests absence of renal failure in rabbits [22].

Table 3 Effect of Pawpaw Seed Oil on the Serum Biochemical Indices of Weaned Rabbits

Variables	T1	T2	Т3	T4	SEM
Albumin (g/dL)	1.92 <sup>b</sup>	2.02 <sup>b</sup>	2.04ª	2.28ª	0.137
Globulin (g/dL)	1.87 <sup>b</sup>	1.92 <sup>b</sup>	2.03ª	2.09ª	0.033
Creatinine (µmol/L)	50.00	51.46	51.49	51.58	0.262
Glucose (Mmol/L)	5.47 <sup>b</sup>	7.04ª	7.65ª	7.21ª	0.231
Urea (Mmol/L)	5.09	5.00	5.17	5.29	0.101

Means in the same row with different superscripts differ significantly (P<0.05); T<sub>1</sub>: basal diet without pawpaw seed oil; T<sub>2</sub>: Basal diet with 0.1 mL pawpaw seed oil; T<sub>3</sub>: Basal diet with 0.2 mL pawpaw seed oil; T<sub>4</sub>: Basal diet with 0.3 mL pawpaw seed oil; SEM: standard error of mean

As presented in Table 3, effect of pawpaw seed oil on the serum enzymes and minerals indices of weaned rabbits. Dietary treatment did not influence (P>0.05) calcium, phosphorus, bicarbonate, alanine phosphatase, aspartate transaminase and alanine transaminase values. Calcium, phosphorus and bicarbonate values were within the normal ranges 1.10 – 3.00 Mmol/L, 0.05 – 2.00 Mmol/L and 1.00 – 4.00 Mmol/L reported by Awosanya et al. when rabbits were fed Pueraria meal[30]. The results suggests the absence of any form of metabolic disorder in rabbits [23]. According to John [31], calcium, phosphorus and bicarbonate are important in maintaining fluid as well as pH balance in the body. Alanine phosphatase, aspartate transaminase and alanine transaminase were within the normal values 60.90 – 91.00 (U/L), 35.00 – 60.00 (U/L) and 25.00 – 45.00 (U/L) cited by Omokore and Alagbe. This result suggests the phyto-constituents in pawpaw seed oil were non-toxic to cause liver damage in rabbits.

Table 4 Effect of Pawpaw Seed Oil on the Serum Enzymes and Minerals Indices of Weaned Rabbits

Variables	T1	T2	Т3	T4	SEM
Calcium (Mmol/L)	2.46	2.50	2.68	2.71	0.06
Phosphorus (Mmol/L)	1.32	1.38	1.41	1.43	0.02

Bicarbonate (Mmol/L)	4.43	4.92	5.01	5.08	0.18
Alanine phosphatase (U/L)	79.71	80.00	80.60	80.72	2.27
Aspartate transaminase	46.31	45.06	45.11	45.18	1.09
(U/L)					
Alanine transaminase	33.05	35.15	36.05	36.17	1.85
(U/L)					

Means in the same row with different superscripts differ significantly (P<0.05);  $T_1$ : basal diet without pawpaw seed oil;  $T_2$ : Basal diet with 0.1 mL pawpaw seed oil;  $T_3$ : Basal diet with 0.2 mL pawpaw seed oil;  $T_4$ : Basal diet with 0.3 mL pawpaw seed oil; SEM: standard error of mean

# **5 CONCLUSION**

In conclusion, pawpaw seed oil contains several phytochemical constituents which possess vital medical significance and pharmacological effects. It is also generally regarded as safe and could serve as alternatives to antibiotics. Pawpaw seed oil can be supplemented up to 0.3 mL/kg diet in the diet of growing rabbits without affecting the health status of animals.

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#### ETHICAL APPROVAL

Animal guidelines was followed in this study for species observation and identification (ANS/0023A).

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## CONFLICT OF INTEREST

The authors have no relevant financial or non-financial interests to disclose.

# DATA AND MATERIALS AVAILABILITY

All data associated with this research are present in the manuscript.

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