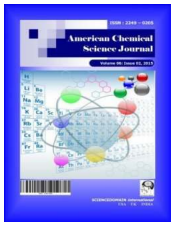

American Chemical Science Journal
15(3): 1-12, 2016, Article no.ACSJ.19789
ISSN: 2249-0205

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Proximate Analysis and Toxicological Studies of *Polyalthia longifolia* Seed Flour in Dietary Formulation of Albino Rats

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Authors' contributions

This work was carried out in collaboration between both authors. Authors IAA and ENI designed the study, performed the statistical analysis, wrote the protocol and wrote the first draft of the manuscript. Authors IAA and ENI managed the analyses of the study. Authors IAA and ENI managed the literature searches. Both authors read and approved the final manuscript.

Article Information

DOI: 10.9734/ACSJ/2016/19789
Editor(s):
(1) Silvia Antonia Brandán, Inorganic Chemistry Institute, National University of Tucumán (UNT), Argentina.
Reviewers:
(1) Manjoosha Srivastava, CSIR- National Botanical Research Institute, Lucknow, Uttar Pradesh, India.
(2) Iyekhoetin Matthew Omoruyi, Benson Idahosa University, Benin City, Edo State, Nigeria.
Complete Peer review History: http://sciencedomain.org/review-history/15303

Original Research Article

Received 26th June 2015
Accepted 15th December 2015
Published 7th July 2016

ABSTRACT

Aim: A short term toxicological study of *Polyalthia longifolia* seed flour in dietary formulation of albino rats was conducted and investigated in order to determine its suitability as an additive in feed supplement and formulation.
Methodology: The effect of the dietary formulation on the physical appearance, feed intake and weight gain of the rats was determined. The proximate analysis, mineral composition, haematological and blood biochemistry were determined and calculated using standard methods of analysis.
Results: The proximate analysis revealed that the seed flour has high carbohydrate and moderate protein contents. The values of 59.66±0.19% for carbohydrate, 12.40±0.25% for protein, 11.90±0.04% for crude fiber, 90.80% for dry matter were obtained for *P. longifolia* seed flour. The seed flour was found to contain high concentration of iron, manganese, zinc, copper and potassium. Other mineral present such as magnesium, calcium and sodium were very low. Nickel, chromium and lead were not present. The low Na/K ratio (0.03) obtained showed that the seed flour could

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probably reduce high blood pressure. Proximate composition of the compounded feed both for the control and the test groups pointed out that there were significance differences in crude protein and crude fibre contents of the feeds compounded. Weekly monitoring of the rats showed good physical appearance and steady weight gain with no mortality recorded during the period of the experiment. The average weight gain per rat at the end of the experiment was shown to be 86.58 g for control rats and 90.00 g for test ones.

Conclusion: Haematological analysis of the rats in both groups showed that they were not anemic and there was no significant difference at p≤0.05 in their blood biochemistry. There was no lesion found in the sections of the kidney, liver and heart of the rats. *P. longifolia* seed flour seemed to be a good supplement in livestock feed formulation.

Keywords: Polyalthia longifolia; chemical analysis; toxicological studies; albino rats.

1. INTRODUCTION

Food crops have occupied an important place in human nutrition as they remain the major sources of calories and proteins for a large proportion of the world population, particularly, in the developing countries [1]. In many of these countries, the supply of animal protein is inadequate to meet the protein needs of the rapidly growing population and the consumption of high amount of meat increases the risk of cardiovascular diseases and some types of cancer. This has necessitated contemporary research that is directed towards studying the food properties and potential utilization of protein from locally available food crops, most importantly from underutilized or neglected high protein oilseeds and legumes [2,3,4]. The nutritional value of any ingredient or feed can be evaluated by biological, chemical and physical scores [5]. Chemical score has proven to be a vital tool in food chemistry because it tends to assess the nutritional value based on the proximate composition of the food and feed which includes the protein, carbohydrate, lipid, moisture, ash and dietary fibre contents respectively. Plants are primary sources of carbohydrate and other nutrients used by human on daily basis. Their roots, leaves, fruits and seeds often provide food for humans [6]. Due to insufficient availability and high cost of animal protein, there has been a constant search for unconventional legumes and oils seed as new source of protein for use as functional supplements in food system [7].

P. longifolia (Sonn.), which belongs to the family of Annonacea*e*, is an evergreen plant commonly used as an ornamental street tree due to its effectiveness in combating noise pollution [8]. The genus *Polyalthia* includes about 120 species occurring mainly in Africa, South and South-Eastern Asia, Australia, and New Zealand. The bark has been reported to be used in the treatment of skin diseases, fever, hypertension, helmenthiasis and in a febrifuge [9]. Its aqueous extract also lowers the blood pressure and rate of respiration in experimental animals [10].

The hypoglycemic and anti-hyperglycemic activity of various solvent extracts of *P. longifolia* leaf extracts was evaluated in alloxan-induced experimental diabetes in rats. *P. longifolia* extracts and powder produced glucose lowering activity, which suggested then that the extracts did not modify any of the biochemical parameter significantly [11]. The antimicrobial activity of the leaves [12], stem [13] and seed diterpenoids [14] have been reported. The plant *P. longifolia* has been reported to contain alkaloids, clerodane di-terpines, quercetin, bulbocapnin, campesterol, diterpenes, and sesquiterpenoids which may contribute to the analgesic activity [15]. *P. longifolia* seed flour, regarded as an under-utilized plant product and with its significant biological and pharmacological activities can be used as additive in the diet formulation. Food additives are substances which are usually included in feeds in trace amounts to preserve its nutritional characteristics prior to feeding (antioxidants and mould inhibitors); facilitate ingredient dispersion or feed pelleting (emulsifiers, binders or stabilizers); promote growth (growth promoters, antibiotics and hormones); facilitate feed intake and acceptance (feeding stimulants and colorants) and also supply some essential nutrients such as minerals and vitamins [16]. Additives improve the nutritional value of certain foods and can make them more appealing by improving their taste, texture, consistency or colour while some modern synthetic preservatives/additives are controversial because they have been shown to cause respiratory or other health problems. Some studies point to synthetic preservatives and artificial coloring agents aggravating symptoms in those affected [17].

We are living in a world where maximum utilization of natural resources is the ultimate goal. Currently, these seeds are not eaten nor used for any industrial purposes in Nigeria. Information on the chemical and nutritional composition of *P. longifolia* seed flour is very scanty while previous workers in the Western part of Nigeria have given information on the phytochemical and fatty acid profile analysis of *P. longifolia* seed oil [18]. There is no reported data on effect of incorporating this seed into the food chain. This paper therefore, reports on the proximate analysis and toxicological study of *Polyalthia longifolia* seed flour on dietary formulation in albino rats. This would provide scientific data regarding their potential application in food processing as well as additive in food supplements.

2. MATERIALS AND METHODS

2.1 Materials

P. longifolia seeds used for this work were collected from Agbowo, Ibadan, Oyo State of Nigeria. The plant seeds were identified and authenticated at the Herbarium Unit of Botany Department, University of Ibadan, Nigeria.

2.2 Physical Characterization

Weight, length and width of the seed and the kernel were determined by taking the average measurement of 20 seeds and kernels. Determinations were done in triplicate. The colour and state of the oil at room temperature were noted by visual inspection.

2.3 Sample Preparation

The fruits were cleaned and decorticated to obtain the seeds. The seeds were dehulled manually, cleaned and separated from the hulls. The kernels were cut into thin slices and then sun-dried at 30±2℃ to constant weight. The dried slices were ground using a mechanical grinder and sieved through a 200 mesh sieve (British standard). The flour obtained was packed in transparent polyethylene bags prior to use.

2.4 Metal Determination

For the metal determination, 0.5 g of each sample was digested with 20 ml of a mixture of concentrated HNO_3 and perchloric acid (2:1 v/v). It decomposes the organic matter completely until the solution becomes a clear one. Thereafter, it was filtered and transferred into a 100 ml standard flask and diluted. It was made up to the mark with deionized water and stored in a clean polyethylene bottle. Sodium and potassium content were determined by using a flame photometer (Model, 405, Corning, UK) as described by Person, (1976), while other mineral element contents were determined using an atomic absorption spectrophotometer (Perkin–Elmer model 703, USA) [19].

2.5 Proximate Analysis

Samples were analysed for dry matter, ash, crude protein, crude fat and total nitrogen according to Association of Official Analytical Chemists methods [20]. Nitrogen content of the seed flour was estimated using the micro-kjeldahl method [21] and the percentage nitrogen was converted to crude protein by multiplying by a factor of 6.25. Carbohydrate contents were determined by difference {100 - (moisture + protein + crude fat + ash + crude fiber)}, [22]. The calorific energy value was obtained by multiplying the values of carbohydrate, protein and crude fat by the Atwater factors of 17, 17 and 37 respectively [23]. Determinations were in triplicate. The crude fat values were used to calculate the total fatty acid by multiplying with a conversion factor of 0.80 [24].

2.6 Feed Compounding

A basal diet was formulated to meet the entire nutrient requirement for young albino rats of 6 weeks (*Rattus norvegius*). *P. longifolia* seed flour was used to totally replace wheat flour in the diet formulated (10%). The diets were prepared according to the procedure described by Souza et al. and Ajayi et al. [25-26] with slight modification. The basic ingredients used are as follows: 35% of maize, 18% of soy bean, 3% of calcium, 1% of salt, 15% of groundnut cake, 7.5% of palm kernel cake, 10% of wheat, 7.5% of corn bran and 3% of oyster shell for the control diet). Wheat was totally replaced with 10% of *P. longifolia* in the experimental diet while 10% of wheat was incorporated in the control diet. Ingredients of the diets were mixed thoroughly with the mixing machine to obtain a homogenous mixture which was pelletized, weighed to be 7000.00 g (100%) for each diet and packed into two different transparent sterile plastic containers for the analysis.

2.7 Animal, Diets and Feeding

Fourteen albino rats (aged 4 - 6 weeks, weighing between 55 g –75 g) were used for this

experiment. They were obtained from the Central Animal House in Veterinary Department, University of Ibadan, Nigeria. The animals were divided into two groups containing seven rats each according to their age gap. They were allowed to acclimatize for one week before the commencement of the experiment. The rats were given feed and water *ad libitum* for an experimental period of 8 weeks before sacrifice. The experimental rats were fed with a compounded feed where wheat was totally replaced with *P. longifolia* flour [26]; the control rats were fed with normal feed compounded with wheat flour alone. The physical appearance of both the control and experimental rats was monitored while the body weight of each rat was recorded weekly (without fasting). At the end of feeding period of eight weeks, the rats were fasted overnight and then sacrificed.

2.8 Collection of Sample from Animals

After the eighth weeks of experiment, the rats were sacrificed under mild anesthetics with chloroform after 14-16 h overnight fast. The blood samples were collected by heart puncture into two heparinized tubes for the studies. One tube contains ethylene diamine tetracetic acid with calcium serving as anti-coagulant in the blood sample for haematological analysis while the second tube was stored at -20℃ for the biochemical studies. Serum alanine aminotransferase and aspartate aminotransferase activity were determined by using commercial kits {Randox laboratories Co Atrium, UK}, [27]. Serum creatinine, urea, total protein, albumin, and globulins and albumin/globulin ratio (A/G) were also determined [28]. The internal organs of the rats (liver, kidney, heart, spleen, lungs, intestine, and brain) were collected, weighed and kept in 10% phosphate buffer formalin for histopathology.

2.9 Haematological Analysis

For haematological analysis, 3 ml of blood were collected by cardiac puncture into heparinized tubes and stored at 10℃ for analysis the same day. The packed cell volume, haemaglobin concentration, red blood cell and white blood cell counts were determined using standard techniques [29]. The differential WBC counts mean corpuscular volume and mean corpuscular haemaglobin concentration were calculated [30-31]. Microhaemocrit capillary tubes were filled to two-thirds mark with well mixed venous blood. One end was sealed with plasticine. The sealed tubes were placed in microhaematocrit centrifuge and the safety cover securely screwed on. The sealed capillary tubes were centrifuged for 5 mins at 10,000 revolutions per minute. The volume of the red blood cell was read on the micro-haemocrit reader.

2.10 Tissue Pathology

A sample of the liver, kidney, heart and spleen for each animal in the various groups was fixed in 10% phosphate buffer formalin, embedded in paraffin wax, sectioned at 6μ and transferred to clean glass slides. The thin sections were stained with haemotoxylin and eosin (H and E) dyes for examination under the light microscope for histological changes in the tissues due to the consumption of *P. longifolia* and seed flour [30].

2.11 Statistical Analysis

Results were expressed as mean±standard deviation. Organ weights, biochemical and hematological determinations were analyzed by student's t-test. A probability level of $p<0.05$ was considered significant.

3. RESULTS AND DISCUSSION

3.1 Physical Characteristics

The physical characteristics of *P. longifolia* seeds with regards to the weight, length and width are listed in Table 1. The average weight of 20 seeds is 21.8±0.03 g. The average length and width of the seed are 2.25±0.18 cm and 1.88±0.10 cm respectively. The kernel percentage of 89.90% was obtained for *P. longifolia* seed.

3.2 Chemical Composition

The chemical composition of *P. longifolia* seed flour was viewed in terms of proximate analysis and mineral composition.

The role of trace elements in nutrition cannot be over emphasized. The mineral elements form a small proportion of the total composition of most plant materials and total body weight but do not contribute to the energy values of food. This elements are still of great physiological importance majorly in the body metabolism [32]. The results for the mineral composition in mg/100 g of *P. longifolia* seed flour (Table 1) show that the seeds have high level of iron (706 mg/100 g), followed by manganese (391 mg/100 g); zinc (342 mg/100 g), copper (342 mg/100 g)

and potassium (102 mg/100 g) for *P. longifolia*. Potassium is an essential mineral element that helps to regulate blood pressure. Calcium, sodium and magnesium were generally low in the seed flours. Nickel, lead and chromium were not detected in the seed flours. The value (0.03) obtained when dividing the concentration of sodium by magnesium for *P. longifolia* is less than 1. This suggests that the seed flour could probably reduce high blood pressure. At the same time, the value (0.05) obtained for Ca/Mg in *P. longifolia* seed flour is very low compared with the recommended ratio of 2.2, National Research Council), [33]. This may be caused by the low calcium content of the flour. It may be important to supplement the seed flour with calcium for diet formulation. The results obtained are favorable compared with that of *Propolis africana* and white melon [34-35].

Table 1. Physical properties and mineral composition of *P. longifolia* seeds

Physical properties	*P. longifolia*
Weight of 20 seeds(g)	21.8±0.03
Weight of a seed (g)	1.09±0.01
Seed length (cm)	2.25±0.18
Seed width (cm)	1.88±0.10
Kernel percentage	89.90%
Colour of unripe seed	Green
Colour of ripe seed	Black
State of the oil @ RT	Liquid
Mineral analysis	**mg/100 g**
Sodium	3.4
Potassium	102.00
Calcium	30.30
Magnesiun	20.90
Manganese	391
Iron	706
Copper	234
Zinc	342
Nickel	ND
Chromium	ND
Lead	ND
NA/K	0.03
Ca/Mg	0.05

The chemical composition of *P. longifolia* seed flour was investigated and presented as shown in Table 2. The moisture content and the dry matter of the seed flour are 9.20±0.03% and 90.80% respectively. These values obtained are in accordance with the values of 11.00±0.34 and 89.00 obtained for wheat flour [36]. The moisture content obtained is higher than 6.93 for pumpkin, 3.48 in bottle gourd and seed flours [37], 5.00 for white melon seed flour [35] and 1.9 for *P. africana* [34]. The ash contents obtained from the seed is 2.58±0.02, values which are lower than 3.91 and 4.4±0.1 obtained for *Lagenaria siceraria* and *P. africana* [23,34]. The crude protein content of *P. longifolia* obtained is 12.40±0.25. This value obtained falls within the range of 8.40-14.8 found for cereal seeds such as corn, triticale and wheat [38]. The value is much lower compared with crude protein in rich foods such as 37.1 in *Lagenaria* seed, [23], 23.6±1.5 in *P. africana* [34], 27.9 in *Parkia biglobos* seed [39] and 36.18±0.04 in moringa seed flour [40]. The carbohydrate content of *P. longifolia* (59.66±0.19) obtained by difference is high. A value of 8.64±0.29 has previously been reported for ginger bread seed flour [41]. *P. longifolia* seeds might be a good source of carbohydrate and it is an indication that they will be good source of roughage in animal feeds. The ash content is quite low while the moisture is high. The calculated metabolic energy value obtained is 1382.64 KJ/100 g for *P. longifolia*. This shows that the seed flour could be a source of energy. Table 3 shows the various energy values as contributed by protein, fat and carbohydrate. The proportion of total energy due to protein is 15.26%, proportion due to fat is 11.39% and proportion due to carbohydrate is 73.35% for *P. longifolia*. This is an indication that the energy obtained from *P. longifolia* seed is majorly contributed by the high carbohydrate content of the seed flours and could be useful as supplement in compounding animal feed.

3.3 Rat Experiment

3.3.1 Effect of *P. longifolia* seed flour on the proximate composition of the compounded feed

After compounding the feed, diet samples were analyzed again to ascertain their chemical composition. It was then observed that there was an increase in the crude protein, ash and crude fat contents in the diet when compared to the pre-diet compounding proximate values. A decrease was also noticed in the moisture and carbohydrate contents while the dry mater value almost remained the same. Interestingly, moisture, crude protein and crude fat contents were not significantly different in both control and test diets groups (Table 3).

Table 2. Proximate composition and energy content of *P. longifolia* seed flour, compounded feed and wheat

Parmeters*	PLSF$	Control group	Test group	Wheat flour#
Moisture content	9.20±0.03[a]	10.15±0.01[a]	8.25±0.01[b]	11.00±0.34
Ash content	2.58±0.02[a]	14.1±0.02[a]	10.07±0.02[b]	1.00±0.04
Crude protein	12.40±0.25[a]	19.23±0.05[a]	19.33±0.12[a]	11.50±0.14
Crude fat	4.26±0.01[b]	15.46±0.01[b]	12.5±0.1[c]	1.50±0.05
Crude fiber	11.90±0.04[a]	7.58±0.14[b]	8.07±0.01[a]	0.6±0.01
Carbohydrate	59.66±0.19[a]	33.46±0.06[b]	41.77±0.12[a]	74.90
Dry matter	90.80	89.89	91.75	89.00
Energy (kj/100 g)	1382.64	1,466.73	1,488.11	1524.3
Calculated fatty acid	3.41	12.37	10.00	1.2

Mean±SD for three replicate analyses, $Values in the same row with the same superscripts are not significantly different at (P< 0.05), #Akurbo et al. (2013)

Table 3. Energy values as contributed by protein, fat and carbohydrate of *P. longifolia* seed flour, compounded feed and wheat

Parameters	PLSF	Control feed	Test feed	Wheat[a]
Total energy	1382.64	1466.73	1488.11	1524.3
Proportion of total energy due to protein %	15.26	22.29	22.08	12.83
Proportion of total energy due to fat %	11.39	39.00	31.08	3.64
Proportion of total energy due to carbohydrate %	73.35	38.78	47.71	83.53

#Akurbo et al. (2013)

Table 4. Effect of *P. longifolia* seed flour on rat body weight and feed gain

Parameters	Control group	Test group
Average initial weight/rat	55.00±11.9	55.00±5.00
Average final weight/rat	141.58±16.18	145.00±17.08
Weight gain/rat	86.58	90.00
% weight gain	157.42	163.63
Survival rate (s) %	100	100

3.3.2 Physical appearance of test and control rats

Weekly inspection of the physical appearance of the rats revealed that the rats were normal in all the groups throughout the eight weeks of the experiment. The eyes, mouth and hair of the animals in both groups appeared to be normal throughout the period of the study. No mortality was recorded for any of the groups.

3.3.3 Body weight and weekly feed consumption of the rats

Figs. 1 and 2 show the weekly feed consumed and the body weight of the different groups of rats. Fig. 1 revealed that the feed consumed increased gradually along the weeks of the experiments. The total feed consumed by the rats in the tests groups are a little higher than those consumed by the rats in the control groups.

In Fig. 2, all the test rats steadily gained weight during the period of the experiment. The average weight gain per rat is shown to be 86.58 g for control and 90.00 g for test group respectively. This indicated that the rats in the test group had a higher average weight gain than those in the control groups.

3.3.4 Organ weights

The organs whose weights were taken are brain, liver, heart, kidney, lungs, spleen and intestine (Fig. 3). At the moment of sacrifice, organs of the test and control rats which are similar have comparable weights; the seed flour has no significant effect ($p<0.05$) on the weight of similar organs of the test and control group of rats. Liver weight was 4.74±0.63 g for the control and 4.62±0.63 g for test. A value of 3.92±0.19 g to 3.95±0.21 g has been previously reported as liver weights for rats fed with crude

Balanite aegyptiaca seed oil in rat [42]. The liver weight of 6.84±1.08 g (control group) to 6.17±1.15 test group) was reported for rat fed with *Greenwayodendron suavelens* seeds [43].

3.4 Haematology, Blood Chemistry and Histopathology

3.4.1 Haematological analysis

There was no significant difference ($p \leq 0.05$) in the result of the haematological analysis of test and control groups (Table 5) of rat fed with *P. longifolia* seed flour. The rats were not anaemic, their PCV values are similar to those reported for healthy murine species [44]. Therefore, the diet compounded with *P. longifolia* seed flour might have no adverse effect on the blood of the rats under this study as it is comparable to the indices obtained for the diet compounded with wheat. This was in accordance with previous result [45-46]. The similarity and the closeness of the WBC counts observed in the control groups when compared to the corresponding test groups suggests that the rats had no infection. This is similar to previous reported work on *T. occidentalis* [47]. The platelets (platelets cells/cu.mm) x 10^6) values obtained were 0.150±0.05 and 0.115±0.03 for control and test groups respectively while the haemoglobin

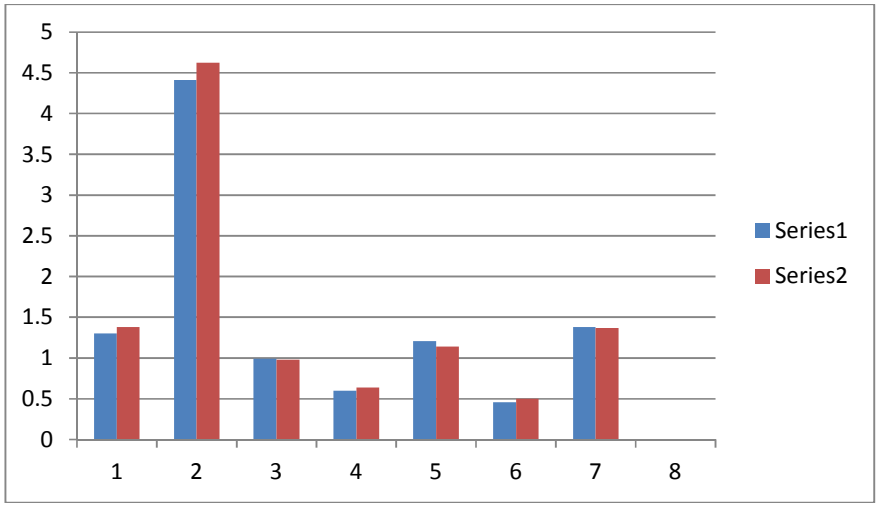

Fig. 1. A chart showing the weekly feed consumption of rats fed with *P. longifolia* seed flour feeds. Series 1 is the control while series 2 is the test group

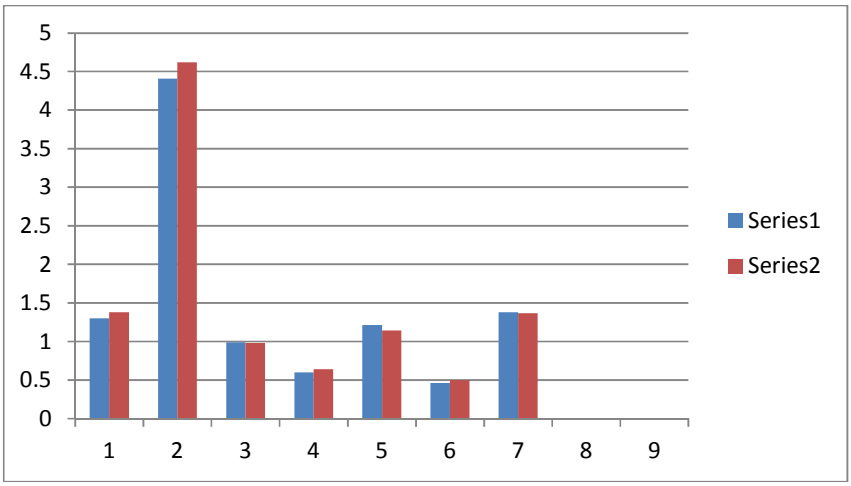

Fig. 2. A chart showing the weekly body weight (g) of rats fed with *P. longifolia* seed flour feed. Series 1 is the control while series 2 is the test group

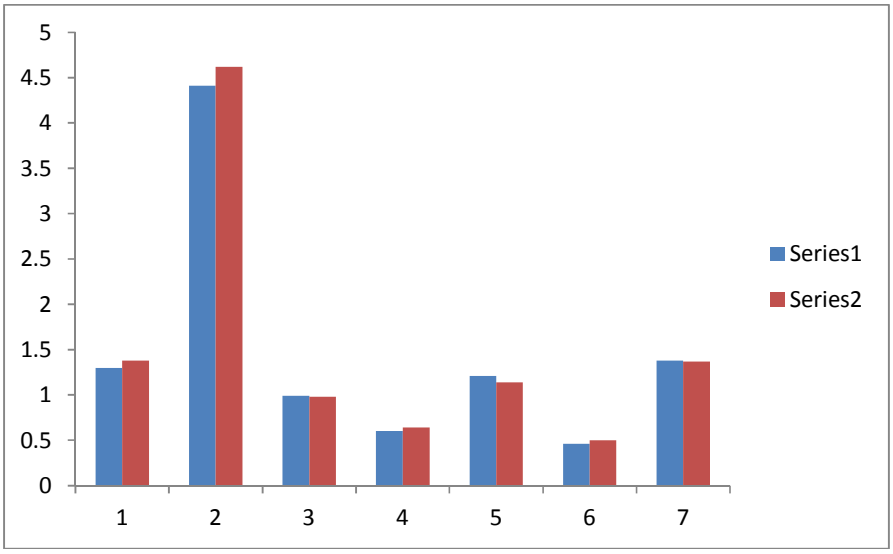

Fig. 3. A chart showing the mean internal organ weight (g) of rats fed with *P. longifolia* seed flour feed. Series 1 is the control while series 2 is the test group. Brain, liver, kidney, heart, lungs, spleen and intestine are represented respectively by 1, 2, 3, 4, 5, 6 and 7 along the y axis

values are 16.73±1.05 and 15.42±1.13 respectively for control and test groups. The haematological values from this study are similar to the result reported on the toxicity study of *Garcinia mangostana* pericarp extract in rats [45].

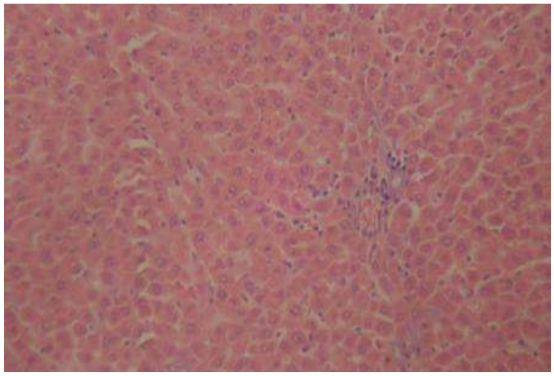

Fig. 4. Photomicrograph of the kidney of test group rats fed with *P. longifolia* seed flour showing no visible lesion. (x550)

3.4.2 Blood chemistry

Table 5 also revealed the result of blood biochemistry of the rats fed with *P. longifolia* seed flour. The serum protein values are comparable to each other in the corresponding control and test groups. Alanine aminotransferases enzyme activity examined was 28.33±0.94 and 30.00± 2.82 respectively for control and test. Alanine aminotransferases enzyme activity value of 20.33±9.87 in the case of the experimental feed as against 15.14±6.41 for the control was previously reported for *Greenwayodendron suavelens* seeds in rat feed [43].

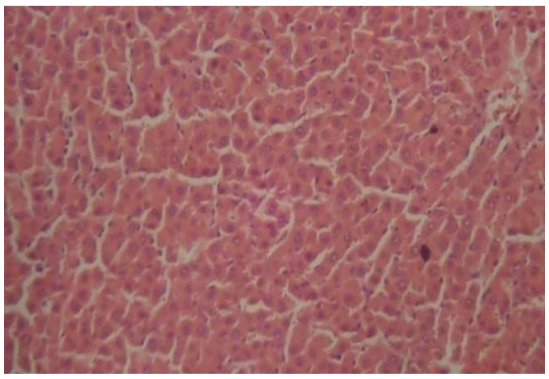

Fig. 5. Photomicrograph of the kidney of control group rats fed with feed without *P. longifolia* seed flour showing no lesion (x550)

3.4.3 Histopathological analysis

No lesion was observed in the heart, kidney and liver of the rats in the control and test groups (Table 6). This is an indication that *P. longifolia* seed flour was probably not harmful to the organs of the rats.

Table 5. Haematological and biochemical studies of rats fed with control feed and *P. longifolia* seed flour

Haematological analysis*	Control group	Test group
PVC (%)	46.86±2.54[a]	46.57±2.82[a]
MCHC (%)	33.11±0.66[a]	34.22±0.36[a]
MCH (%)	20.1±0.53[a]	23.42±0.05[a]
MCV (%)	60.72±1.48[a]	62.21±8.33[a]
Hb (mg/de)	16.73±1.05[a]	15.42±1.13[a]
RBC (10^6/ml)	8.14±0.60[a]	7.68±0.62[a]
WBC (10^3/ml)	8,793±0.79[a]	7,607±2.9[a]
Lymplocyte (%)	79.001±9.35[a]	72.00±5.38[a]
Neutrophil (%)	20.57±8.83[a]	23.14±3.74[a]
Monocyte (%)	2.43±1.27[a]	2.57±1.72[a]
Eosinophil (%)	2.00 ±1.15[a]	2.86±3.83[a]
Platelets (cells/cu.mm)x10^6	0.150±0.05[a]	0.115±0.03[a]
Biochemical analysis*		
TP (g/dl)	7.86±0.30[a]	6.77±0.70[a]
ALB (g/dl)	5.07±0.22[a]	4.07±0.60[a]
GLB (g/dl)	2.33±0.60[a]	2.67±0.11[a]
AL/GLB	2.17±0.57[a]	1.52±0.2[b]
AST (g/l)	40.00±1.00[a]	41.33±2.43[a]
ALT (g/l)	28.33±0.94[a]	30.00±2.82[a]
ALP (g/l)	80.33±6.67[a]	73.33±1.15[a]
Urea	14.67±0.71[a]	15.00±1.00[a]
Creatinine	0.73±0.1[a]	0.6±0.10[a]

Hb = Haemoglobin, concerntration (g%); PCV = Packed cell volume (%), RBC = Red Blood Cell Counts, WBC = White Blood cell count (x10^3/mm3), MCV = Mean Corpuscular Volume (fl), MCH = Mean Corpuscular Haemoglobin (%); MCHC = Mean Corpuscular Haemoglobin Concentration (%). AST- Aspartate aminotransferases, ALT- Alanine aminotransferases, ALP = Alkaline phosphatase; ALB = Albumin; GLB = Globulin; ALB/GLB = Albumin – Globulin ratio; TP = Total Protein

***Values in the same row with the same superscripts are not significantly different at (P< 0.05)*

Table 6. Histological result of tissues from the rats fed with *P. longifolia* seed flour

Tissue	Control group	Test group
Kidney	No visible lesion	No visible lesion
Liver	No visible lesion seen	No visible lesion seen
Heart	No visible lesion	No visible lesion

4. CONCLUSION

P. longifolia seed flour could be utilized successful as source of dietary supplement in feed for livestock due to the protein content and the high carbohydrate values. Their low protein content can still be supplemented with other high protein residue such as groundnut cake, soy bean cake, *B. aegyptiaca* cake and melon cake. *P. longifolia* seed flour is very high in iron, manganese, zinc, copper and potassium but low in calcium, sodium and magnesium. Therefore, it might be important to supplement the seed flour with calcium for diet formulation.

P. longifolia seed flour, when fed to rat, did not have any deleterious effect on the physical appearance, body weight gain, organ weights, haematological and biochemical parameters. The seed flour was found not to be toxic to the heart and kidney of the rats. No lesion was seen in the liver of the rat fed with *P. longifolia.* It is then suggested that the seed flour might be a good source of nutrient for livestock diet formulation. *P. longifolia* seed flour can still be studied at graded level of incorporation in order to determine the suitable level or percentage at which the seed flour could possibly and effectively be integrated in formulating feed for livestock.

COMPETING INTERESTS

Authors have declared that no competing interests exist.

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