HAEMATOLOGICAL AND SERUM BIOCHEMICAL CHARACTERISTICS OF WEANER PIGS FED GRADED LEVELS OF BAMBARA GROUNDNUT OFFAL

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ABSTRACT
A 56 day study was conducted using 12 crossbred weaner pigs of average weight 10.67kg to determine the effects of different levels of raw bambara groundnut (Vigna subterranea) offal on the haematology and serum chemistry of the pigs. The 12 animals were divided into four groups of 3 each, and randomly assigned to four treatments in a completely randomized design (CRD). The pigs were housed in individual pens, with each animal representing a replicate. The raw BGO was included in the diets at 0, 10, 20 and 30% levels. Feed and water were offered ad-libitum. Blood was collected from the ear lobe of each animal at the end of the study period. The parameters measured were haemoglobin (Hb), packed cell volume (PCV), white blood cell (WBC), red blood cell (RBC), neutrophils, lymphocyte, monocyte and eosinophil and serum chemistry, which included, glucose, urea, creatinine, total bilirubin, conjugated bilirubin, total protein, aspartate amino transferase (AST) and alanine aminotransferase (ALT). The results showed that the red blood cell (RBC×10⁶µl), haemoglobin (Hb), packed cell volume (PCV %), white blood cell (WBC×10³µl), neutrophils (%) and lymphocytes (%) differed (P<0.05) significantly among the treatment diets. The values were, red blood cell 4.75, 4.55, 3.80 and 4.70 x10⁶µl, haemoglobin 12.70, 10.10, 9.25, and 10.05 g/dl, packed cell volume 37.00, 30.50, 28.00 and 30.50 %, white blood cell 10.20, 15.95, 14.00 and 19.90 x10³µl, neutrophils 43.00, 35.00, 36.00 and 24.50% and lymphocytes 52.50, 62.00, 60.50 and 62.00 % for diets 1, 2, 3 and 4 respectively. Glucose, urea (mg/dl) and total protein (g/dl) differed (P<0.05) significantly among the treatment means. The values were, glucose 18.30, 14.60, 10.05 and 11.00 mg/dl, urea 62.35, 42.25, 45.30 and 52.40 mg/dl and total protein 6.00, 8.70, 6.65 and 5.00 g/dl) for diets 1, 2, 3 and 4 respectively. However, monocyte, eosinophil, creatinine, total bilirubin, conjugated bilirubin, aspartate aminotransferase and alanine aminotransferase did not differ (P>0.05) significantly among the treatment means. The result indicated that raw bambara groundnut offal can be incorporated into diet for weaner pigs at 30% level of inclusion without adverse effect on the physiological welfare of the animals.

Keywords: Feeding, bambara groundnut, offal, weaner, pigs and haematology

INTRODUCTION
Nigerian pig industry is facing tremendous setback and on the verge of collapse arising from high cost of pig feed which accounts for 70-80% of the total cost of pig production in Nigeria and other developing countries (Olomu, 1995). The escalating market prices of cereal grains especially maize which forms the bulk of pig diet is highly competitive and not readily available because of the high demand by human as staple food and it’s industrial use. However, the high cost of conventional feed stuff most especially the protein supplement, necessitated the quest for locally available alternatives that can substitute for the conventional feed stuffs economically by reducing feeding cost, thereby making the pig enterprise more profitable. In order to address this increase in the price of conventional feed stuffs and produce pigs and pig products of affordable prices in Nigeria, the use of agro-industrial by-products must be exploited in pig feeding.
There is evidence in literature that the use of agro-industrial by products reduce the cost of feeding as they attract little pricing (Onyimonyi, 2002; Onuh, 2005, Okah and Onwujiariri, 2010). A large number of alternative feedstuffs which have potentials as monogastric feed ingredients abound in Nigeria (Ologbogbo,1992). These include industrial by-products and Agricultural wastes like maize offal, maize milling waste, soyabean hulls, cowpea hull, cashew nut offal, bambara groundnut offal, etc. However, there are no reliable production statistics for those by-products because of scattered processing centers (Fetuga and Tewe, 1994). Moreover poor industrial and technological base of the country results in fragmental supply of these materials and this makes standardization for physical quality or nutrient content difficult (Ukachukwu, 2000). Feed producers using these categories of ingredients can hardly maintain standards. These by-products have no direct feeding value for human, and in most places it is dumped indiscriminately thus, constituting environmental hazards.

Bambara groundnut (*Vigna subterranea*) has been reported to be good source of plant protein that is useful for feeding of chicks (Olomu, 1995). In Nigeria, most of the bambara groundnut produced are consumed locally. No industrial use of the crop has been reported (Tanimu and Aliyu, 1995). A survey conducted in the Southeastern part of Nigeria shows that it is abundant, and the machines which are used to grind the nuts before they are sieved into flour for various uses are also available in almost all the local markets. This has led to increased availability of the offal (waste) obtained after sieving out the flour. The proximate composition of seed has been reported by Olomu (1995) as 20.6%CP; 6.3%EE; 4.0%CF; 4.2%Ash and 54.9%NFE, although these vary with different processing methods as have been reported by Igbodioh *et al.* (1994). Okah *et al.* (2006) reported the proximate composition of raw and autoclaved bambara nut as; 17.71%CP,3.05%CF,13.30%EE,3.15%Ash and 49.29%NFE for raw bambara nut, and 16.83%CP,4.10%CF,12.40%EE,3.25%Ash and 50.67%NFE for autoclaved bambara nut.

Pig production in particular represents one of the fastest ways of increasing animal protein intake, since pigs grow at a faster rate and are highly prolific than cattle, sheep and goat, in growth rate, only surpassed by broilers (Holness, 2005). Apart from their high rate of reproduction, pigs are characterized by the best efficiency of nutrient conversion into high quality animal protein (Smith, 2001; Holness, 2005). On an average pigs now produce 100kg gain from 300-350kg feed, i.e. a kilogram of pork can be produced on as little as 3.0-3.3 kilogram of feed (Banerjee, 2005). Different animal species and breeds have their different ranges of haematological and serum biochemical characteristics. However, haematological and serum biochemistry characteristics of animals are generally influenced among other factors by the quality of the feeds consumed by such animals. This study was conducted to evaluate the haematological and serum biochemistry of weaner pigs fed graded levels of bambara groundnut offal.

**MATERIALS AND METHODS**  
**The experimental site**  
The experiment was conducted at the piggery unit of the Teaching and Research farm of College of Animal Science and Animal Production (CASAP), Michael Okpara University of Agriculture, Umudike, Umuahia, Abia State; located at latitude 5° 29′ north and longitude 1.7° 32′ east in the rain forest zone of Nigeria. The experiment was conducted within July and August 2013. The climate of the region is characterized by a mean daily temperature of between 27°C and 35°C all through the year. Average rainfall of the place is about 2000mm per annum with double maxima pattern (NRCRI, 2013).  
**The experimental animal and design**  
The weaner pigs for the experiment were commercial breed bought from the Michael Okpara University of Agriculture commercial pig farm, umudike, Umuahia, Abia State. Twelve (12) crossbred
weaner pigs of Large White × Landrace which were eight (8) weeks of age were used to conduct a 56-day experiment in a Completely Randomized Design (CRD) experiment. There were four (4) treatments and three (3) animals per treatment, each animal representing a replicate.

The statistical model is as follows:

\[ X_{ij} = \mu + T_i + e_{ij} \]

\[ X_{ij} \] = single observation
\[ \mu \] = overall mean
\[ T_i \] = the \( i^{th} \) treatment effect
\[ e_{ij} \] = Error term

**Procurement of feed ingredients**
The bambara groundnut offal was sourced from the local millers at the Umuahia main market. Other feed ingredients were also obtained from JOCAN AGRO LTD, Umuahia. Samples of the raw bambara groundnut offal and diets were analysed for proximate composition (A.O.A.C., 2012). Four (4) experimental diets were formulated for weaner pigs (table 1).

**Table 1: Composition of experimental diets containing**

<table>
<thead>
<tr>
<th>Ingredients</th>
<th>( T_1 )</th>
<th>( T_2 )</th>
<th>( T_3 )</th>
<th>( T_4 )</th>
</tr>
</thead>
<tbody>
<tr>
<td>Maize</td>
<td>39.80</td>
<td>32.80</td>
<td>25.80</td>
<td>18.80</td>
</tr>
<tr>
<td>SBM</td>
<td>18.00</td>
<td>15.00</td>
<td>12.00</td>
<td>9.00</td>
</tr>
<tr>
<td>BGO</td>
<td>0.00</td>
<td>10.00</td>
<td>20.00</td>
<td>30.00</td>
</tr>
<tr>
<td>Maize offal</td>
<td>12.00</td>
<td>12.00</td>
<td>12.00</td>
<td>12.00</td>
</tr>
<tr>
<td>PKC</td>
<td>27.00</td>
<td>27.00</td>
<td>27.00</td>
<td>27.00</td>
</tr>
<tr>
<td>Bone meal</td>
<td>2.50</td>
<td>2.50</td>
<td>2.50</td>
<td>2.50</td>
</tr>
<tr>
<td>Salt</td>
<td>0.25</td>
<td>0.25</td>
<td>0.25</td>
<td>0.25</td>
</tr>
<tr>
<td>Vit/Min Premix*</td>
<td>0.25</td>
<td>0.25</td>
<td>0.25</td>
<td>0.25</td>
</tr>
<tr>
<td>Lysine</td>
<td>0.10</td>
<td>0.10</td>
<td>0.10</td>
<td>0.10</td>
</tr>
<tr>
<td>Methionine</td>
<td>0.10</td>
<td>0.10</td>
<td>0.10</td>
<td>0.10</td>
</tr>
<tr>
<td>Total</td>
<td>100.00</td>
<td>100.00</td>
<td>100.00</td>
<td>100.00</td>
</tr>
</tbody>
</table>

**Calculated Composition**

<table>
<thead>
<tr>
<th>CP (%)</th>
<th>( \text{ME(Kcal}^{-1})</th>
<th>( \text{Kg} )</th>
<th>( \text{Kg} )</th>
<th>( \text{Kg} )</th>
<th>( \text{Kg} )</th>
</tr>
</thead>
<tbody>
<tr>
<td>17.83</td>
<td>3026.75</td>
<td>3020.15</td>
<td>3013.57</td>
<td>3006.99</td>
<td></td>
</tr>
</tbody>
</table>

**Different levels of raw bambara nut offal**

SBM= Soybean meal; BGO= Bambara nut offal

*supplied per kg diet: Vit. A 7500IU; Vit. D 750; Vit. K 3mg; Thiamine 10mg; Riboflavin 5mg; Niacin 20mg; Pantothenate 15mg; Vit. B 22mg; Biotin 50mg; Choline 300mg; Magnesium 500mg; Iodine 0.20mg; Zinc 100mg; Iron 90mg; Manganese 20mg; Selenium 0.15mg; methionine 0.10% and Lysine 0.10%.

**Collection of Data**
The Haematological parameters measured included; packed cell volume (PCV), Haemoglobin (Hb), Red blood cell count (RBC), White blood cell count (WBC), Neutrophils, Lymphocytes and Eosinophils. The biochemical parameters included; Creatinine, Total bilirubin, Total protein, Conjugated bilirubin, Glucose, Urea, Aspartate aminotransferase (AST) and Alanine aminophosphatase (ALP).
Blood Collection and Analysis

The blood samples were collected from one animal per treatment using sterilized needles and syringes. The collection was done in the morning and 10mls of blood was obtained from the ear lobe of the animal into two sample vaccutainer tubes, one containing ethylene diamine tetra-acetic acid (EDTA) for haematological study and the other sterile vaccutainer tube without EDTA for serum chemistry. The Haematological parameters; packed cell volume (PCV), Haemoglobin (Hb), Red blood cell count (RBC), White blood cell count (WBC), Neutrophils, Lymphocytes and Eosinophils were analyzed according to Baker et al. (1998). Serum metabolites (serum protein, albumin, bilirubin, urea, glucose) were determined using the methods described by Toro and Ackerman (1975). While AST and ALT, were determined according to Rej and Hoder (1983). All data generated were subjected to one way analysis of variance (ANOVA) according to Steel and Torrie (1980), and where significant differences existed, Duncan’s multiple range test (Duncan, 1955) were used to separate the means using SAS (1999).

RESULTS AND DISCUSSION

The analysed composition of the experimental diets containing different levels of raw bambara nut offal and BGO is represented in table 2. The crude protein content of 12.43% is lower than 17.90% reported by Amaefule and Osuagwu (2005), 17.17% (Okah et al., 2006). The ether extract of 3.27% reported in this study is lower than 13.30% (Okah et al 2006) and 6.30% (Olomu,1995). The crude fiber of 13.00% obtained for raw BGO in this study is also higher than the 11.30% reported by Amaefule and Osuagwu (2005) and 5.40% by Amaefule and Iroanya (2004). The ash content (3.11%) of BGO reported herein is similar to the 3.15% (Okah et al., 2006). The metabolizable energy values (MJkg⁻¹) of the T2 (10%raw BGO) (1.67) and T4(30%raw BGO) (1.68) were higher (p<0.05) than that of the control (T1, 0% raw BGO) (1.57), while the T1 and T3 ( 20% raw BGO) (1.63) were similar (p>0.05). However, the 57.38% NFE for raw BGO reported herein is higher than the 49.29% (Okah et al., 2006) and 54.90% (Olomu, 1995).

Table 2: Analyzed composition of the experimental diets containing different levels of raw bambara nut offal and BGO

<table>
<thead>
<tr>
<th>Constituents</th>
<th>$T_1$</th>
<th>$T_2$</th>
<th>$T_3$</th>
<th>$T_4$</th>
<th>SEM</th>
<th>BGO</th>
</tr>
</thead>
<tbody>
<tr>
<td>Dry Matter (%)</td>
<td>89.73a</td>
<td>89.66a</td>
<td>89.38b</td>
<td>89.43b</td>
<td>0.094</td>
<td>89.19</td>
</tr>
<tr>
<td>Crude Protein (%)</td>
<td>9.98d</td>
<td>13.83a</td>
<td>11.90c</td>
<td>12.95b</td>
<td>0.212</td>
<td>12.43</td>
</tr>
<tr>
<td>Ether Extract (%)</td>
<td>4.56a</td>
<td>2.84c</td>
<td>4.60a</td>
<td>4.32b</td>
<td>0.037</td>
<td>3.27</td>
</tr>
<tr>
<td>Crude Fibre (%)</td>
<td>3.94d</td>
<td>6.20c</td>
<td>6.53b</td>
<td>7.25a</td>
<td>0.037</td>
<td>13.00</td>
</tr>
<tr>
<td>Ash (%)</td>
<td>8.71a</td>
<td>3.41d</td>
<td>7.41b</td>
<td>4.09c</td>
<td>0.008</td>
<td>3.11</td>
</tr>
<tr>
<td>NFE (%)</td>
<td>62.54b</td>
<td>63.38a</td>
<td>58.94d</td>
<td>60.82c</td>
<td>0.272</td>
<td>57.38</td>
</tr>
<tr>
<td>ME(MJkg⁻¹)*</td>
<td>1.59b</td>
<td>1.67a</td>
<td>1.63ab</td>
<td>1.68a</td>
<td>0.021</td>
<td>1.68</td>
</tr>
</tbody>
</table>

Means within the same row with different superscript differ significantly (P<0.05). NFE – Nitrogen Free Extract,
*Calculated

The bambara groundnut offal is obtained by manual sieving of milled bambara groundnut while separating the flour for human consumption. The extent of sieving, amount of residual flour, particle size and the sieving skill of the miller affect the quality or the proximate composition of bambara groundnut offal. These factors could lead to variations among bambara groundnut offal obtained from different millers, and over a period of time. The crude protein values of the raw BGO diets were higher (p<0.05) than that of the control diet, but variation among the raw BGO diets was due to different ratios of the raw BGO to soybean meal (SBM) in the diets. Higher (p<0.05) ether extract value in the
control diet (T₁) might be due to higher oil content of SBM compared to raw BGO. However, the variation among the BGO diets might be due to different ratios of SBM: BGO. The crude fibre content of diets increased with level of BGO in the diet due to high fibre content of the BGO used in the experiment. The NFE (%) differed significantly (p<0.05) among the treatment diets without a definite pattern, and appeared to have been due to varying ratios of the variable ingredients (maize, SBM and BGO).

Table 3 represents the haematological, while table 4 shows the serum biochemical characteristics of weaner pigs fed graded levels of bambara nut offal.

Table 3: Haematological characteristics of weaner pigs fed with graded level of bambara nut offal

<table>
<thead>
<tr>
<th>Parameters</th>
<th>T₁</th>
<th>T₂</th>
<th>T₃</th>
<th>T₄</th>
<th>SEM</th>
</tr>
</thead>
<tbody>
<tr>
<td>Hb (g/dl)</td>
<td>12.70a</td>
<td>10.10b</td>
<td>9.25b</td>
<td>10.05b</td>
<td>0.55</td>
</tr>
<tr>
<td>PCV (%)</td>
<td>37.00a</td>
<td>30.50b</td>
<td>28.00b</td>
<td>30.50b</td>
<td>1.66</td>
</tr>
<tr>
<td>WBC (×10³ µl)</td>
<td>10.20d</td>
<td>15.95b</td>
<td>14.00c</td>
<td>19.90a</td>
<td>0.42</td>
</tr>
<tr>
<td>RBC (×10⁶ µl)</td>
<td>4.75</td>
<td>4.55</td>
<td>3.80</td>
<td>4.70</td>
<td>0.18</td>
</tr>
<tr>
<td>Neutrophil (%)</td>
<td>43.00a</td>
<td>35.50b</td>
<td>36.00b</td>
<td>24.50c</td>
<td>0.87</td>
</tr>
<tr>
<td>Lymphocyte (%)</td>
<td>52.50b</td>
<td>62.00a</td>
<td>60.50a</td>
<td>62.00a</td>
<td>2.18</td>
</tr>
<tr>
<td>Monocyte (%)</td>
<td>0.00</td>
<td>0.00</td>
<td>0.00</td>
<td>1.00</td>
<td>0.00</td>
</tr>
<tr>
<td>Eosinophil (%)</td>
<td>2.00</td>
<td>1.00</td>
<td>1.00</td>
<td>1.20</td>
<td>0.00</td>
</tr>
</tbody>
</table>

a,b,c Means in rows with different superscript indicates significant difference (p<0.05).

Hb = Haemoglobin, PCV = Packed cell volume, WBC = White blood cell, RBC = Red blood cell

Table 4: Serum biochemical characteristics of weaner pigs fed diets containing graded levels of bambara nut offal

<table>
<thead>
<tr>
<th>Parameters</th>
<th>T₁</th>
<th>T₂</th>
<th>T₃</th>
<th>T₄</th>
<th>SEM</th>
</tr>
</thead>
<tbody>
<tr>
<td>Glucose(mg/dl)</td>
<td>18.30a</td>
<td>14.60b</td>
<td>10.05c</td>
<td>11.00c</td>
<td>0.24</td>
</tr>
<tr>
<td>Urea(mg/dl)</td>
<td>62.35a</td>
<td>42.25c</td>
<td>45.30c</td>
<td>52.90b</td>
<td>2.87</td>
</tr>
<tr>
<td>Creatinine (mg/dl)</td>
<td>1.00</td>
<td>0.80</td>
<td>0.80</td>
<td>0.90</td>
<td>0.33</td>
</tr>
<tr>
<td>Total Bilirubin(mg/dl)</td>
<td>0.60</td>
<td>0.60</td>
<td>0.60</td>
<td>0.70</td>
<td>0.35</td>
</tr>
<tr>
<td>Conc. Bilirubin(mg/dl)</td>
<td>0.20</td>
<td>0.20</td>
<td>0.20</td>
<td>0.20</td>
<td>0.00</td>
</tr>
<tr>
<td>Total protein(g/dl)</td>
<td>6.00b</td>
<td>8.70a</td>
<td>6.65b</td>
<td>5.00c</td>
<td>0.00</td>
</tr>
<tr>
<td>AST (µ/l)</td>
<td>7.00</td>
<td>6.00</td>
<td>7.00</td>
<td>7.00</td>
<td>0.00</td>
</tr>
<tr>
<td>ALT (µ/l)</td>
<td>4.00</td>
<td>3.50</td>
<td>3.00</td>
<td>4.00</td>
<td>0.00</td>
</tr>
</tbody>
</table>

a,b,c Means in rows with different superscript indicates significant difference (p<0.05).

AST (SGOT) = Aspartate aminotransferase, ALT (SGPT) = Alanine aminotransferase

The hemoglobin (Hb) and packed cell volume (PCV) were significantly (p<0.0%) lower in the animals fed BGO diets than in those fed the control diet. However, the Hb and PCV values which ranged from 9.25 to 10.10g/dl and 28.00 to 30.50% respectively for the BGO diets were similar (p>0.05). Decrease in Hb and PCV is associated with the presence of toxic factors in diets such as hemaglutinin (Oyawole and Ogunkunle (1998). The presence of anti nutritional factors such as trypsin inhibitors, polyphenol have been reported (Poulter, 1981), hemaglutinin, tannins, cynonogenic glucosides and flatulence
factors in raw bambara nut have been reported (Enwere, 1998). It is possible, that the presence of anti
nutrients in the raw BGO may have depressed erythropoiesis but did not cause hemolysis as there was
no anemic condition in the pigs. White blood cell (WBC (x10^3 µl) recorded higher (p<0.05) values (T2
(15.95), T3 (14.00) and T4 (19.90) in the animals fed BGO diets than in those fed the control T1
(10.20). Although the values of WBC in the animals fed the BGO diets were higher, they were still
within the normal range of 11-22 (x10^3 µl) (Banergee, 2005) for pigs. Neutrophil (%) level was higher
(p<0.05) in the group fed control diet than those fed raw BGO diets. The lymphocyte (%) levels of the
experimental animals indicated higher (p<0.05) values for animals fed diets containing raw BGO than
those fed the control diet. The reduction in neutrophil and increase in levels of lymphocyte are
indicative of immunologic responses resulting from the stimulation of WBC due to ingestion of some
levels of the anti nutrients in the diets containing raw BGO. The monocyte (%) and eosinophil (%) were
not influenced by the treatment. The biochemical characteristic of the animals fed diets containing
graded levels of raw BGO is presented in table 4. The serum metabolites indicated significantly
(p<0.05) higher level of glucose in the control group (T1 = 18.30mg/dl) than the raw BGO groups, T2
(14.60mg/dl), T3 (10.05mg/dl) and T4 (11.00mg/dl. Higher glucose level in the group fed the control
diet T1 (0%BGO) suggests that there was more readily fermentable carbohydrates in that diet than the
BGO diets as reported by Okah and Ibeawuchi (2011) that higher serum glucose indicated more
readily fermentable carbohydrate in the diet. Blood urea (mg/dl) was significantly (p<0.05) higher in
the control group than in the BGO groups. However the values were within the normal range as
reported by Ologhobo and Fetuga, (1993) High blood urea level is an indication of poor protein quality
(Egun, 1970) or poor protein metabolism (Oyawoleand Ogunkunle, 1998). It therefore follows that the
raw BGO diets were of higher protein quality or were better metabolized than the control diet as the
animals that consumed them recorded lower blood urea. Total serum protein was significantly (p<0.05)
higher in the group that were fed the T2 (10% raw BGO) with the value 8.70mg/dl. The groups fed T1 (0% raw
BGO) and T3 (20% raw BGO) had similar (p>0.05) serum protein levels of 6.00 and 6.65mg/dl respectively.
The amount of dietary protein appears to be a major but not an exclusive indicator of serum protein level. The
physiological status and nutrients interactions might also influence serum protein and other metabolites. The
dietary treatments did not influence the creatinine which was within the normal range reported by Ross et al.,
(1978), total and conjugated bilirubin, and metabolic enzymes, aspartate aminotranferase (AST) and alanine
aminotransferase (ALT) whose presence in the serum indicate muscle damage as myocardial infarction.

CONCLUSION
The haematological and serum chemistry of weaner pigs fed varying dietary levels of raw bambara nut
offal (BGO) did not show any adverse effect on the physiological status of the animals. Therefore, 30% raw
BGO could be included in the diet of weaner pigs, as the result of this study indicated that the pigs tolerated it.
Further studies might be necessary with higher inclusion levels of raw BGO to establish the optimal level of
inclusion of raw BGO for weaner pigs and other production stages of pigs. There is currently high pressure
already on the available bambara groundnut as human food and its waste as animal feed resource, making its
supply inadequate. Therefore, increased production of the nut must be encouraged by Government and
stakeholders in the agricultural sector to bridge the gap of inadequate supply and sustain its contribution in the
national economy.

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