HAEMATOLOGICAL AND SERUM BIOCHEMICAL RESPONSES OF WEST AFRICAN DWARF GOATS FED *PANICUM MAXIMUM* REPLACED WITH UNTREATED COCOA POD HUSK MEAL

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ABSTRACT

It has been proven that blood profile of an animal dictates its health status. Meanwhile, the health implication of feeding graded levels of untreated cocoa pod husk meal (UCPHM) to animals was the crux of this study. Thus, a 56 day trial was conducted to assess the haematological and biochemical responses of West African Dwarf (WAD) goats fed Panicum maximum replaced with UCPHM. The feed components were blended into five diets such that Diet A (control) contained 100 % P. maximum, Diet B (75 % P. maximum and 25 % UCPHM), Diet C (50 % P. maximum and 50 % UCPHM), Diet D (25 % P. maximum and 25 % UCPHM) while Diet E (100% UCPHM) and were fed to 25 WAD goats of 5 replicates per treatment in a completely randomized design experiment. The dry matter (DM), crude protein (CP), crude fibre (CF) and gross energy (GE) contents of the diets were adequate to support the growth and maintenance of WAD goats. The values obtained for parameters observed, except CP increased with increased replacement for UCPHM across the treatment diets. Nutrient intake and apparent digestibility co-efficient values were significantly (p<0.05) influenced by dietary treatment. WAD goats fed with Diet A had better haematological profiles, though the RBC, WBC, lymphocyte and neutrophil values had statistically (p>0.05) similar values with goats fed Diet B (75 % P. maximum + 25 % UCPHM). Hence, replacing P. maximum with untreated cocoa pod husk at 25 % replacement level was acceptable and tolerable to the WAD goats and would not pose any health challenges.

Keywords: Haematology, Serum biochemical, Untreated cocoa pod husk, *Panicum maximum,* West African dwarf goat

INTRODUCTION

Nutrition in livestock plays a germane role in the overall productivity and the health status of the animals as feed accounts for the major proportion (60 – 70 %) of the total cost of intensive animal production (Abiola *et al*., 2012). Its insufficiency in supply in terms of the quality and quantity may bring about a condition referred to as malnutrition in farm animals which will consequently result to low productivity in such deprived animals.

It is therefore imperative for farmers or animal producers to give more attention to animal nutrition if maximum productivity is to be attained. This, therefore calls for the search of alternative feed resources such as agro-industrial waste and crop residue that could be harnessed to improve and sustain animal performance and productivity to meet up with the animal protein (especially from goat) demand by human being. World production of cocoa (*Theobroma cacao*) beans, which is primarily for the manufacture of chocolate and

cocoa powder, currently stands at 4.05 million metric tonnes (ICCO, 2012). Cocoa production is important to the economy of Nigeria, it is the leading agricultural export of the country and Nigeria was the third largest cocoa producer in Africa after Ghana (WCF, 2012). Hence, the by-product (cocoa pod husk) is been generated in huge quantities, and it is been regarded as a waste as its use in livestock feeding is still underutilized. Cocoa pod husk had been nutritional reported to contain between the range of 80 – 88.96 % dry matter (DM), 6 - 9.14 % crude protein (CP), 24.93 - 35.74 % crude fibre (CF) and 14.1 - 21.16 % lignin (Sobamiwa and Longe, 1994; Iyayi *et al*., 2001; Alemawor *et al.,* 2009).

Guinea grass (*Panicum maximum*) is highly productive, palatable, nutritive and can be solely fed to livestock or fed alone with concentrates or legumes, and has proven to be excellent forage for ruminant animals (Bamikole *et al*., 2001).

Blood is useful for assessing the health status, clinical evaluation for survey of physiological and pathological conditions, diagnostic and prognostic evaluation of various types of diseases in animals (Etim *et al.,* 2013). However, there is paucity of information on haematological and serum biochemical responses of WAD goats fed *P. maximum* replaced with untreated cocoa pod husk meal (UCPHM). Thus, this study provides information on the effects of graded levels of UCPHM on the blood profile of WAD goats.

MATERIALS AND METHODS

Experimental Site: The experiment was conducted at Teaching and Research Farm (Small Ruminant Unit) of the Federal University of Technology, Akure (Latitude 7^0 15″N and Longitude 5^0 15″E) (Ajibade *et al.,* 2014), Ondo State, Nigeria.

Ingredients Collection and Preparation: Cocoa pod husks were collected from Odudu Cocoa Plantation at Ilara-mokin, Akure, Ondo State. The cocoa pod husks were sun-dried for 10 days and crushed at the University feed mill into 1 mm diameter size. *P. maximum* were harvested at the University farm, air dried for a day and chopped to smaller sizes before feeding.

Preparation of Experimental Diets: Five experimental diets were formulated such that Diet A (control) which contained 100 % *P. maximum*; Diet B (75 % *P. maximum* and 25 % UCPHM), Diet C (50 % *P. maximum* and 50 % UCPHM); Diet D (25 % *P. maximum* and 75 % UCPHM) and Diet E (100 % UCPHM).

Laboratory Procedures: Proximate analyses were carried out on the experimental diets according to AOAC (2002) method.

Moisture content determination: This was done using the oven-drying method. Cleaned, dry and well-labelled Petri dishes were weighed (W_1). About 5 g each of the samples were weighed into the dishes (W_2) and transferred into the oven maintained at 105 °C for 3 hours. After three hours, they were transferred into the desiccators to cool and then weighed (W_3). This process continued every hour until a constant weight was obtained. Percentage moisture = loss in weight of sample (W_3 - W_2) ÷ weight of samples before drying (W_2 - W_1) x 100.

Determination of crude protein: This involved three stages namely; digestion, distillation and titration.

Digestion: About 0.5 g of sample was weighed into 500 ml Kjeldahl flask. 10 ml of concentrated H_2SO_4 and selenium catalyst were added; and boiled until the sample turned into a clear solution. It was cooled and made up with distilled water to 50 ml. The sample was stored in a bottle.

Distillation: 5 ml of 2 % H_2BO_3 (boric acid) was added into the conical flask and 2drops of mixed indicator (0.198 g bromocresol green plus 0.132 g methyl red into 200 ml alcohol) were added. The 5ml of digested sample was pipetted into the condenser's cup and l0ml of 40 % NaOH was added, this was washed with distilled water. The joints were tightened and

distillation was done till a volume of 50 ml was reached in the receiving flask.

Titration: The distillate was titrated with 0.1 ml HCl until the end point (pink colour) was reached. Percentage nitrogen = Titre value x 0. 1M HCl x 0.014 x 100x V_1/V_2 ÷ weight of sample x 100. Where, V_1 = volume of digest (50 ml), V_2 = volume of digest used (5 ml). Percentage crude protein = % nitrogen x 6.25.

Determination of crude fibre: One gram of the sample was weighed into the conical flask (W_1) and boiled with 1.25 % H_2SO_4 (150 ml) for 30 minutes. The mixture was filtered through a poplin cloth and rinsed with distilled water; the sample was then scraped back into the flask and boiled with 150 ml of 1.25 % NaOH for 30 minutes. The residue was collected again and rinsed with distilled water, 10 % HCl and lastly with ethanol. The residue was later scrapped into a crucible, oven dried, cooled in the desiccator, weighed (W_2) and ashed in the muffle furnace. It was removed into desiccator and allowed to cool to room temperature, then re-weighed (W_3). Percentage crude fibre = weight after oven drying (W_2) - weight after removal from furnace (W_3) ÷ weight of sample (W_1) x 100.

Determination of ether extract: Filter paper was weighed (W_1) and 0.5 g of each of the oven dried samples were added into it and weighed (W_2). The filter paper with the content was neatly folded, tied using thread arranged in the thimble. Round bottom flask (500 ml capacity) was filled with n-hexane up ¾ of the flask. The extractor was fitted with the reflux condenser and heated to allow the solvent boil gently; and siphon several times within 4 hours. Samples were then removed, dried in the oven for one hour at 105 °C, cooled in the desiccators and weighed (W_3). Percentage ether extract (fat) = loss in weight of the sample (W_2- W_3) ÷ weight of dried sample (W_2-W_1) x 100.

Determination of ash: Dry and clean crucibles were weighed (W_1) and their respective weights recorded, about 1g of the samples were added and weighed again (W_2). The crucibles and contents were placed into the muffle furnace at 600^0C until a light grey colour of ash was obtained. The crucibles were removed and allowed to cool in the desiccators and then weighed (W_3). Percentage ash = weight of ash (W_2- W_3) ÷ weight of sample (W_2 W_1) x 100.

Nitrogen free extract (NFE): This refers to the soluble carbohydrate in the sample and it is obtained by difference. Percentage NFE = 100 - (% ash + % crude fibre + % crude protein + % fat + % moisture).

Energy determination: The gross energy (GE) of the diets was determined by the methods of Eknayake *et al*. (1999) as follows: GE (KJ/l00g DM) = % CP x 16.7 + % EE x 37.7 + % CHO x 16.7. Where, CHO = carbohydrate, CP = crude protein and EE = ether extract.

Determination of Theobromine Concentration: Two grams of the sample was weighed into a glass stopper 500 ml bottle followed by adding 270 ml chloroform and 10 ml ammonia solution (10 % m/m NH_3), shake vigorously for 5 minutes. 12 grams of anhydrous sodium sulphate was added to the solution, shake again and allowed to stay overnight. The solution was washed and filtered with 100 ml chloroform. The solvent was distilled off while the trace of chloroform was removed by heating in an oven at 100 ^{o}C; 50 ml of water was added to the residue and boiled. The solution was later neutralized with 0.1 M sodium hydroxide solution using 0.5 ml of 1 % ethanol solution of phenol red as indicator. 20 ml of 0.1 M silver nitrate solution was also added. The liberated acid was titrated with 0.1 M sodium hydroxide to the red colour in the indicator to determine the level of theobromine. 1 ml of NaOH = 18.0 mg theobromine. Percentage theobromine = titre valve x 18.0 x 100 ÷ sample weight x 1000.

Experimental Layout: Twenty-five (25) West African dwarf goats with average live-weight of 7.15 ± 0.07 kg and 12 – 18 months old were selected from the goat flock of Teaching and Research Farm of FUTA. The goats were

vaccinated against *Peste-Petit-de Ruminante* (PPR / kata) using PPR vaccine at the rate of 1ml per animal, treated against endo and ectoparasites using Ivomectin and against infections by using oxytetracycline LA (long acting) at the 1 ml per 10 kg body weight of animal. The animals were randomly distributed into five treatment groups of five goats per replicate using the completely randomized experimental design.

An acclimatization period of fourteen days (after quarantine) was allowed before commencement of data collection. Animals were fed 5 % of their body weight early in the morning (8:00 am) and supplied cool, fresh drinkable water (*ad libitum*) during the experimental period of 56 days. Feed left unconsumed were weighed and discarded. The daily feed intake was determined by deducting the refusals from the quantity offered. The animals were weighed before the commencement of experiment and were repeatedly weighed weekly in the morning before feeding, to observe any weight change using spring-balance (hanging scale).

Total faeces were collected in the morning before feeding and watering during last 7 days of the experiment. The faeces were weighed fresh and 10 % of faeces collected from each animal were taken and oven dried at 105 ^{0}C for 48 hours to determine the nutrients of the faeces according to AOAC (2002) procedures. Apparent digestibility coefficient of the diets was calculated as difference between nutrients intake and excretion in the faeces, expressed as a percentage of nutrient intake.

Blood Collection: Blood samples were collected via the jugular vein puncture (Frandson, 1986) before and at the end of the experiment from each animal to analyze for both haematological and biochemical components. Blood samples were taken into separate bottles containing anticoagulant (ethylene diamine-tetra-acetate EDTA) and the other with anticoagulant free bottles from which serum was harvested for biochemical analysis.

Haematology: Packed cell volume (PCV) was determined by collecting the blood samples containing anticoagulant and rocked gently for thorough mixing and drawn up using micro-haematocrit capillary tubes to 3/4 of its length. One end of the tube was sealed with plasticine. The capillary tube was then placed in micro haematocrit centrifuge with plasticine end outward. The tubes were centrifuged at 1,200 rpm for 4 minutes and then read with haematocrit reader. The reading expressed the PCV as a percentage of the total blood volume according to Thrall and Weiser (2002). Haemoglobin concentration determination was done according to Higgins *et al.* (2008) using the calorimetric method which involved a haemoglobin standard. One gram of pure haemoglobin was ground in porcelain mortal and 0.1 g of it was weighed and dissolved in 10 ml of distilled water using a glass rod to stir continuously until the haemoglobin had been thoroughly dissolved. This solution was the haemoglobin standard. Each of the blood samples (0.02 ml) were placed in a labelled test tube containing 4 ml of Drabkins solution using an automatic filling pipette. To completely flush out the blood, Drabkins solution was repeatedly poured into the pipette and ejected until a complete flushing was observed. The test tubes were covered, content thoroughly mixed using an automatic spin mixer and left to stand for 5 minutes to allow full colour development. A standard solution was prepared using the same way of haemoglobin standard. The calorimeter was switched on and allowed to warm for about 15 minutes before setting at zero and wavelength set at 624 nm to read the samples. A blank solution was inserted and full scale zero was set. The standard dilution was read and recorded before each sample was inserted one after the other and their corresponding readings recorded. Red Blood Cell count, white blood cells and differential white blood count (lymphocyte, neutrophil, monocyte, eosinophil and basophil) were assayed according to Thrall and Weiser (2002) method.

Serum Biochemistry: The serum indices were carried out using commercial test kits, Quimica Clinica Aplicada (QCA) test kits (QCA, Spain) and a digital colorimeter (Lab-tech, India). The serum total protein was determined

by direct biuret method (Lubran, 1978) and the serum albumin was determined by Bromocresol green method (Doumas *et al.*, 1971). The serum globulin was calculated as the difference between serum total protein and serum albumin (Colville, 2002). The serum alanine aminotransferase (ALT) and aspartate aminotransferase (AST) activities were assayed by the method of Reitman and Frankel (1957). The serum alkaline phosphatase (ALP) activity was determined by phenolphthalein monophosphate method (Babson *et al.*, 1966).

Statistical Analysis: The data collected were subjected to Analysis of Variance using SAS (2008) and significant means were separated using Duncan New Multiple Range (Duncan, 1955).

RESULTS

The percentage proximate composition of the experimental diets was presented in Table 1. The DM content of the experimental diets increase with increased replacement of UCPHM and varied from 88.20 ± 0.14 % (Diet A) to 89.47 ± 0.13 % (Diet E). The crude protein content decreased with increased replacement of UCPHM, the highest value was recorded for diet A (11.28 ± 0.10 %) and diet E had the least value (10.37 ± 0.08 %). Consequently, CF content of the experimental diets increased with increased replacement of UCPHM and ranged from 28.76 ± 0.40 % (Diet A) to 33.10 ± 0.31 % (Diet E). Theobromine concentration was significantly different ($p<0.05$) across the dietary treatment and varied from 0.05 ± 0.03 % (Diet A) to 0.93 ± 0.09 % (Diet E), while the gross energy (GE) also increased across the treatments with increased replacement of UCPHM.

The nutrients intake and apparent digestibility co-efficient by WAD goats fed experimental diets was presented in Table 2. Dry matter intake (DMI), crude protein intake (CPI), CF intake (CFI) and nitrogen free extract intake (NFEI) values were statistically similar ($p>0.05$). The DMI decreased with increased replacement of UCPHM and varied from 341.38 ± 0.19 g/day (Diet A) to 162.69 ± 0.05 g/day (Diet E). The CPI varied significantly ($p<0.05$) across the treatment, but decreased with increased replacement levels of UCPHM and ranged from 43.65 ± 3.01 g/day (Diet A) to 18.87 ± 2.29 g/day (Diet E). The CFI was highest in goats fed Diet B (113.10 ± 3.55 g/day). Goats fed Diet A had the highest (138.65 ± 0.10 g/day) and those fed Diet E had the least (55.50 ± 0.05 g/day) value of NFEI. Theobromine intake was highest in goats fed Diet E (1.70 ± 0.17 g/day) and least in those fed Diet A (0.15 ± 0.09 g/day). The gross energy intake was found to be highest in animals fed Diet D (66.04 ± 4.01 g/day) and lowest in animals fed Diet E (38.78 ± 1.50 g/day).

The dry matter digestibility varied from 57.91 ± 1.62 % (Diet E) to 73.84 ± 1.63 % (Diet A). The crude protein digestibility was highest in goats fed Diet A (81.31 ± 1.30 %). The CF digestibility ranged from 84.30 ± 0.87 % (Diet E) to 90.74 ± 0.75 % (Diet A). The ether extract digestibility ranged from 53.32 ± 2.98 % (Diet E) to 72.41 ± 2.35 % (Diet A) while the NFE digestibility ranged from 55.53 ± 3.14 % (diet E) to 71.31 ± 2.61 % (Diet A). Theobromine digestibility decreased with increased replacement of UCPHM and the gross energy digestibility was highest in goats fed Diet A (80.37 ± 2.47 %) and least value was recorded for those fed Diet E (65.43 ± 0.01 %).

The serum biochemical parameters of WAD goats fed experimental diets (Table 3) revealed that all the parameters were significantly influenced ($p<0.05$) by the dietary treatments. The total protein in goats fed Diet A had the highest value (43.21 ± 3.01 g/l), while those fed Diet E had the least value (17.36 ± 1.19 g/l). The albumin content reduced with increased level of cocoa pod husk meal which ranged from Diet A (25.03 ± 1.84 g/l) to Diet E (8.45 ± 1.62 g/l). Goats placed on Diet B had significantly high ($p<0.05$) globulin value (18.44 ± 0.69 g/l), while those placed on Diet E had the least value (8.91 ± 1.16 g/l). Aspartate aminotransferase was highest in goats fed Diet E (78.91 ± 1.89 IU/l) and least in goats fed Diet A (60.50 ± 1.27 IU/l). Goats placed on Diet E had the highest alkaline phosphatase levels (30.41 ± 3.54 IU/l), while the least value was occurred in goats fed Diet A (10.12 ± 0.83 IU/l).

Table 1: Proximate composition of experimental diets fed to WAD goats

Nutrients	Diets (%)				
	A	B	C	D	E
Dry matter	88.20±0.14^{b}	88.35±0.13^{b}	88.85±0.13^{b}	89.16±0.12^{a}	89.47±0.13^{a}
Crude protein	11.28±0.10^{a}	11.06±0.10^{a}	10.74±0.09^{b}	10.57±0.08^{b}	10.37±0.08^{b}
Crude fibre	28.76±0.40^{e}	29.86±0.42^{d}	30.93±0.39^{c}	32.02±0.53^{b}	33.10±0.31^{a}
Ether extract	2.20±0.33^{e}	3.21±0.39^{d}	4.26±0.46^{c}	5.34±0.42^{b}	6.42±0.40^{a}
Ash	10.71±0.21^{a}	10.33±0.19^{b}	9.74±0.16^{c}	9.33±0.14^{d}	8.74±0.25^{e}
Nitrogen free extract	35.82±0.42^{a}	34.33±0.45^{b}	33.28±0.41^{c}	31.73±0.62^{d}	30.53±0.60^{e}
Theobromine	0.05±0.03^{a}	0.35±0.04^{b}	0.54±0.09^{c}	0.74±0.10^{d}	0.93±0.09^{e}
Gross energy (KJ/100gDM)	13.35±0.75^{e}	15.37±0.72^{d}	17.35±0.73^{c}	19.26±0.73^{b}	21.33±0.70^{a}

a, b, c, d, e = means within the same row with different superscripts are significantly different (p<0.05)

Table 2: Nutrient intake and digestibility co-efficient by WAD goats fed experimental diets

Parameters	Diets				
	A	B	C	D	E
Intake (g/day)					
Dry matter	341.38± 0.19^{a}	334.60±0.11^{a}	316.97±0.21^{ab}	305.77±0.29^{b}	162.69±0.05^{c}
Crude protein	43.65±3.01^{a}	41.90±1.99^{a}	38.30±0.78^{b}	36.25±2.11^{b}	18.87±2.29^{c}
Crude fibre	111.31±6.68^{a}	113.10±3.55^{a}	110.33±6.17^{a}	109.81±5.62^{a}	60.20±5.83^{b}
Ether extract	8.52±0.65^{d}	12.17±0.90^{c}	15.19±0.91^{b}	18.32±1.00^{a}	11.68±0.99^{c}
Ash	41.46±2.41^{a}	39.10±3.21^{a}	34.73±2.33^{b}	32.03±1.94^{c}	15.89±2.31^{d}
Nitrogen free extract	138.65±0.10^{a}	130.04±0.08^{ab}	118.74±0.09^{b}	108.91±0.08^{c}	55.50±0.05^{d}
Theobromine	0.15±0.09^{a}	1.31±0.18^{b}	1.94±0.19^{d}	2.54±0.25^{e}	1.70±0.17^{c}
Gross energy	51.68±1.64^{c}	58.22±2.79^{b}	61.91±3.16^{ab}	66.04±4.01^{a}	38.78±1.50^{d}
Digestibility (%)					
Dry matter	73.84±1.63^{a}	67.01±1.79^{ab}	65.97±1.80^{abc}	61.63±1.81^{c}	57.91±1.62^{d}
Crude protein	81.31±1.30^{a}	80.63±0.96^{a}	77.67±0.98^{b}	76.10±0.89^{b}	72.97±0.71^{c}
Crude fibre	90.74±0.75^{a}	87.97±0.56^{a}	86.62±0.91^{ab}	85.32±0.71^{b}	84.30±0.87^{c}
Ether extract	72.41±2.35^{a}	68.26±2.84^{ab}	63.91±2.97^{b}	61.81±2.91^{b}	53.32±2.98^{c}
Nitrogen free extract	71.31±2.61^{a}	70.14±2.54^{a}	67.13±2.53^{ab}	65.52±2.83^{b}	55.53±3.14^{c}
Theobromine	83.64±2.97^{a}	75.45±2.81^{ab}	72.40±2.40^{b}	66.51±2.69^{bc}	58.00±3.78^{c}
Gross energy	80.37±2.47^{a}	72.04±1.61^{b}	71.62±1.00^{b}	65.84±3.01^{c}	65.43±0.01^{c}

a, b, c, d, e = means within the same row with different superscripts are significantly different (p<0.05)

Table 3: Serum biochemical indices of WAD goats fed experimental diets

Parameters	Diets				
	A	B	C	D	E
Total protein (g/l)	43.21±3.01^{a}	39.66±2.57^{a}	35.47±2.51^{b}	26.61±3.47^{c}	17.36±1.19^{d}
Albumin (g/l)	25.03±1.84^{a}	21.22±1.45^{b}	17.08±1.73^{c}	12.16±1.51^{d}	8.45±1.62^{e}
Globulin (g/l)	18.18±1.12^{a}	18.44±0.69^{a}	18.39±0.14^{a}	14.46±2.54^{a}	8.91±1.16^{b}
Albumin : Globulin	1.40±0.29^{a}	1.19±0.01^{ab}	0.93±0.02^{ab}	0.87±0.02^{b}	0.99±0.01^{ab}
Aspartate aminotransferase (IU/l)	60.50±1.27^{d}	69.96±1.73^{c}	72.68±1.91^{bc}	74.45±1.75^{b}	78.91±1.89^{a}
Alkaline phosphatase (IU/l)	10.12±0.83^{d}	11.94±0.91^{d}	17.67±1.89^{c}	22.25±2.88^{b}	30.41±3.54^{a}
Alanine transferase (IU/l)	41.60±3.52^{c}	42.60±2.67^{c}	47.53±3.86^{c}	62.20±3.49^{b}	76.27±5.01^{a}

a, b, c, d, e = Means on the same row but with different superscripts are significantly different (p<0.05)

The alanine transferase increased with increased in the replacement of UCPHM, the highest value was observed in goats fed with Diet E (76.27 ± 5.01 IU/l), while the least value was observed in goats fed Diet A (41.60 ± 3.52 IU/l). The haematological variables of WAD goats before and after the trial were presented in Table 4. There were no significant difference ($p>0.05$) in all the basic blood profile before feeding the experimental diets to the WAD goats. However, significant differences ($p<0.05$) existed in all the blood profile after feeding the experimental diets to the WAD goats. The PCV decreased with increased replacement level of UCPHM and ranged from 33.00 ± 1.56 % (Diet A) to 18.13 ± 1.35 % (Diets E).

Table 4: Haematological parameters of WAD goats before and after feeding the experimental diets

Parameters	Time	Diets				
		A	B	C	D	E
Packed cell volume (%)	F	24.50±0.35	23.50±0.12	27.00±0.01	27.25±3.75	26.75±0.92
	T	33.00±1.56^{a}	30.00±1.49^{b}	26.67±0.94^{c}	21.67±2.01^{d}	18.13±1.35^{e}
Red blood cell (×10^{6} µl)	F	9.49±0.21	8.68±0.41	9.93±0.22	11.52±0.01	10.93±2.25
	T	13.14±0.14^{a}	12.05±0.95^{a}	9.92±0.86^{b}	9.73±0.94^{b}	6.96±0.11^{c}
White blood cell (×10^{3} µl)	F	3.78±0.01	3.49±0.05	3.93±0.04	4.33±0.55	4.23±0.20
	T	4.44±0.22^{a}	4.05±0.14^{ab}	3.85±0.15^{b}	3.59±0.01^{bc}	3.14±0.13^{c}
Haemoglobin (g/dl)	F	8.18±0.20	7.83±0.21	8.98±0.01	9.10±1.27	8.93±0.01
	T	11.03±0.52^{a}	10.00±0.49^{b}	9.13±0.36^{b}	8.00±0.32^{c}	7.00±0.31^{d}
Lymphocyte (%)	F	63.00±0.25	62.50±0.25	63.75±0.26	63.00±0.12	63.00±0.12
	T	63.33±1.64^{a}	62.00±1.42^{a}	60.00±1.52^{a}	54.67±1.53^{b}	48.67±1.54^{c}
Neutrophil (%)	F	25.25±0.27	25.75±0.36	25.00±0.33	25.00±0.34	25.50±0.35
	T	31.67±1.11^{a}	29.00±0.98^{a}	24.33±0.99^{b}	24.00±0.90^{b}	24.00±0.92^{b}
Monocyte (%)	F	7.50±0.22	7.50±0.24	6.75±0.25	7.50±0.24	7.75±0.25
	T	2.67±0.47^{d}	3.33±0.44^{cd}	4.67±0.46^{bc}	6.00±0.60^{ab}	6.67±0.23^{a}
Eosinophil (%)	F	3.50±0.01	3.50±0.01	3.75±0.23	3.75±0.25	3.25±0.25
	T	3.33±0.02	3.33±0.06	3.33±0.04	3.67±0.25	4.00±0.67
Basophil (%)	F	0.75±0.10	0.75±0.10	0.75±0.09	0.75±0.25	0.50±0.01
	T	0.67±0.16	0.67±0.18	1.00±0.14	1.33±0.12	1.33±0.10

a, b, c, d, e = means within the same row with different superscripts are significantly different (P< 0.05). F= before feeding the experimental diets, T= after feeding the experimental diets

The RBC also decreased across the treatment with an increased replacement of UCPHM level which ranged from 6.96 ± 0.11 x 10 ^{6}µl (Diet E) to 13.14 ± 0.14 x 10 ^{6}µl (Diet A). The WBC ranged from 4.44 ± 0.22 x 10^{3}µl (Diet A) to 3.14 ± 0.13 x 10^{3}µl (Diet E). The haemoglobin level was least in animals fed Diet E (7.00 ± 0.31 g/dl), although goats fed with diets B and C had statistically similar (p>0.05) values. Goats fed Diet A had the highest value of lymphocyte (63.33 ± 1.64 %), while those fed Diet E had the least (48.67 ± 1.54 %). The neutrophil value ranged from 24.00 ± 0.92 % (Diet E) to 31.67 ± 1.11 % (Diet A), although Diets D and E were statistically similar (p>0.05). There was increased in the monocyte value with increased replacement of UCPHM level which ranged from 2.67 ± 0.47 % (Diet A) to 6.67 ± 0.23 % (Diet E). The value of the eosinophil ranged from 3.33 ± 0.02 % (Diet A) to 4.0 ± 0.67 % (Diet E), although Diets A, B and C were statistically similar (p>0.05). Likewise, the value of basophil increased with increased replacement level of UCPHM which ranged from 0.67 ± 0.16 % (Diet A) to 1.33 ± 0.10 % (Diet E), although Diets A and B were statistically similar (p>0.05), also, Diets D and E were statistically similar (p>0.05).

DISCUSSION

The high DM values could be attributed to the nature/texture of the cocoa pod husk (dried), its maturity at harvest coupled with the stage of cutting the guinea grass which also contributed to the dry matter content of the diets and this agreed with the report of Alemawor *et al*. (2009) who reported 88.96 % DM for dried cocoa pod husk. The CP content of the diets decreased with increased substitution of UCPHM and this implied that the nitrogen contribution from the grass enhanced the protein content of the diets. Meanwhile, the nitrogen free extract of the diets had a positive influence on the crude protein of the diets. The recorded dietary crude protein contents were adequate to support growth in ruminants as this will enact the activities of the rumen microbes. The gradual increasing values of CF content in the diets might be attributed to the high lignocellulosic content in the feed materials (*P. maximum* and dried UCPHM). The ether extract content also increased with substitution of cocoa pod husk meal and this had a positive reflection on the energy contents of the diets.

The theobromine is more concentrated in diet E and because of the presence of alkaloid, making it bitter and unpalatable for the goats. Thus, could be the cause of reduced feed intake observed.

The crude protein content of the diets influenced diets intake positively, meanwhile the WAD goats fed diets A and B recorded similar values. The reduction in the dry matter intake could be traced to the bitter taste of cocoa pod husk. Perhaps, this could be responsible for the gradual reduction in feed/nutrient intake as the UCPHM increases in the diets. The gross energy intake by the goats showed the quantity at which they ate to satisfaction and likewise their tolerance level. The recorded values showed that the UCPHM is the major determinant for the DM and gross energy of the diets. This corroborated the assertion of Alexander *et al*. (2008) who reported that cocoa pod had low protein content, high crude fibre and the inherent anti-nutrient (theobromine – an alkaloid substance which has slightly bitter taste and poisonous to animals), would reduce feed/nutrient intake.

It was observed that the nutrient digestibility decreases as UCPHM increases in the diets. The better nutrients digestibility coefficient values recorded for goats fed diets A and B could be traced to the quantity of CP intake by WAD goats and this agreed with the report of McDonald *et al.* (1995) that there is a positive relationship between digestibility of feed and protein intake. The CF digestibility values increased as the crude protein content of the diet increased which correlates with the observation of Ranjhan (1993) who reported that higher protein intake may increase the digestibility of the CF of the feed and this is because of the activity of the microorganisms which are increased on high protein ration and consequently, they attack the CF more vigorously. The bulkiness of the diets C, D and E as a result of increased replacement of UCPHM could be responsible for the reduced digestibility of the diets.

Before feeding the experimental diets, the baseline of the goats showed no variation in all the experimental goats before the trial and no indication of ill-health as all the variables were in line with values reported for healthy goats (Plumb, 1999). The effects of the diets on the haematogical parameters are good indicators of the physiological status and therefore, they are always a reflection of animal responsiveness to their internal and external environment (Etim *et al.,* 2013).

It was observed that the PCV, WBC, RBC, Hb values gradually reduced with increased replacement levels of UCPHM in the diets and the resultant low values in goats fed diets C, D and E showed that the animals would have less defense ability against any infection as a result of low WBC. However, high value of WBC has been associated with toxicity of diets or poor detoxification process which lead to increased production of WBC to fight foreign substances in the body, while low value suggest susceptibility to infection (Nwakolor, 2001). However, this might be the reason for diseased condition (central nervous system depression, restlessness, diarrhoea, muscle tremor, ataxia, anaemia, emaciation and seizures) and the mortality recorded in these treatments which agreed with the report of Alexander *et al.* (2008).

The goats fed diets A and B had improved RBC values which fell within the normal values of 8 – 18 x 10^6 ul/l) recommended by Plumb (1999) for healthy goats. This indicated that the feed was of high quality, not protein deficient, digestible and had tolerable level of antinutrients. Plumb (1999) reported that low level of eosinophils indicated no allergic reactions. Although, from this study the eosinophil values recorded were within the normal range (1 – 8 %) for healthy goats, indicating that there was no allergic reaction imposed by the dietary treatments, but morbidity and mortality were recorded at UCPHM replacement above 25 %.

The statistical similar values of parameters assessed in sera of goats fed diets A and B showed the nutritive potentials of 25 % replacement of UCPHM in this present study. This indicated that the diets were of better quality and supported the physiological function of goats fed, hence their better performance. Awojobi and Opiah (2000) observed that the higher the value of globulin, the greater the

ability to fight infection because globulin is known to fight infection. The measurement of the AST level is helpful for the diagnosis of cases of myocardial infarction and skeletal muscle disorders (Jain, 1993). Perhaps, this implied that, goats fed diets C, D and E had severe liver injury and this could be responsible for the nervous disorder and infections. High value of alkaline phosphatase may have been as a result of the poor protein quality of the experimental diets C, D and E.

Conclusion: From the foregoing, it could therefore be concluded that combining guinea grass and UCPHM at 3:1 would not have any deleterious effect on their health status of WAD goats especially during off-season of the grass. From this study, it is therefore recommended that (i) Diet B (75 % *P. maximum* + 25 % UCPHM) can be adopted by goat farmers to encourage utilization of cocoa pod husk which is still been regarded as a waste in Nigeria, (ii) livestock (goats) farmers should be sensitized about the nutritional potentials of cocoa pod husk and its tolerance level because of the presence of its major antinutrient (theobromine) and (iii) treatment/processing of cocoa pod husk should be to encourage for effective utilization by WAD goats.

REFERENCES

ABIOLA, S. S., RADEBE, N. E. and UMESIOBI, D. O. (2012). Whole hatchery waste meal as alternative protein and calcium sources in broiler diets. *Archivos de Zootecnia,* 61(234): 229-234.

AJIBADE, F. O., ADEWUMI, J. R. and OGUNTUASE, A. M. (2014). Design of improved storm water management system for the Federal University of Technology, Akure. Nigerian *Journal of Technology,* 33(4): 470 – 481.

ALEMAWOR, F., DZOGBEFIA, V. P., ODDOYE, E. O. and OLDHAM, J. H. (2009). Enzyme cocktail for enhancing poultry utilisation of cocoa pod husk. *Scientific Research and Essays,* 4(6): 555 – 559.

ALEXANDER, J., BENFORD, D., COCKBURN, A., CRAVEDI, J. P. and DOGLIOTTI, E. (2008). Theobromine as undesirable substances in animal feed: scientific opinion of the panel on contaminants in the food chain. *The European Food Safety Authority Journal,* 72(5): 61 – 66.

AOAC (2002). *Official Methods of Analysis*. 17th Edition, Association of Official Analytical Chemists, Washington DC, USA.

AWOJOBI, H. A. and OPIAH, G. O. (2000). The effect of physiological status on some blood parameter of the New Zealand white doe rabbits. Pages 14 – 16. *In*: *Proceedings of Animal Science Association of Nigeria.*

BABSON, A. L., GREELEY, S. J., COLEMAN, C. M. and PHILIPS, G. E. (1966). Phenolphthalein monophosphate as a substrate for serum alkaline phosphatase. *Clinical Chemistry*, 12: 482 – 490.

BAMIKOLE, M. A., EZENWA, I., AKINSOYINU, A. O., ARIGBEDE, M. O. and BABAYEMI, O. J. (2001). Performance of West African dwarf goats fed Guinea grass–Verano stylo mixture, N-fertilized and unfertilized Guinea grass. *Small Ruminant Research*, 39(2): 145 – 152.

COLVILLE, J. (2002). Blood chemistry. Pages 75 – 103. *In*: HENDRIX, C. M. (Ed.). *Laboratory Procedures for Veterinary Technicians.* 4th Edition, St. Louis, Mosby, USA.

DOUMAS, B. T., WATSON, W. A. and BIGGS, H. G. (1971). Albumin standards and the measurement of serum albumin with bromocresol green. *Clinica Chimica Acta*, 31(1): 87 – 96.

DUNCAN, D. B. (1955). Multiple range and multiple F tests. *Biometrics*, 11(1): 1 – 42.

EKNAYAKE, S., JANSZ, E. R. and NAIR, B. M. (1999). Proximate composition, mineral and amino acid content of mature *Canavalia gladiata* seeds. *Food Chemistry*, 66(1): 115 – 119.

ETIM, N. N., ENYENIHI, G. E., WILLIAMS, M. E., UDO, M. D. AND OFFIONG, E. E. A. (2013). Haematological parameters: indicators of the physiological status of

farm animals. *British Journal of Science*, 10: 33 – 45.

FRANDSON, R. D. (1986). *Anatomy and Physiology of Farm Animals*. Lea and Fabiger, Philadelphia, USA.

HIGGINS, T., BEUTLER, E. and DOUMAS, B. T. (2008). Measurement of haemoglobin in blood. Pages 514 – 515. *In*: BURTIS, C. A., ASHWOOD, E. R. and BRUNS, D. E. (Eds.), *Tiez Fundamentals of Clinical Chemistry*, 6th Edition, Saunders Elsevier, Missouri, USA.

ICCO (International Cocoa Organization) (2012). Production of cocoa beans. *Quarterly Bulletin of Cocoa Statistics*, 37(4): http://www.icco.org/about-us/ international-cocoa-agreements/catview/30-related-documents/46-statistics-production.html. Accessed 06 December, 2012.

IYAYI, E. A., OLUBAMIWA, O., AYUK, A., OROWVEGODO, S. and OGUNAIKE, E. F. (2001). Utilization of urea treated and untreated cocoa pod husk based diets by growing pigs: An on-farm study. *Tropicultura,* 19(3): 101 – 104.

JAIN, N. C. (1993). *Essentials of Veterinary Haematology.* Lea and Fabiger, Malvern, Philadelphia, USA.

LUBRAN, M. M. (1978). The measurement of the serum total proteins by the Biuret method. *Annals of Clinical and Laboratory Science,* 8(2): 106 – 110.

MCDONALD, P., EDWARD, R. A., GRENHAIGH, J. F. D. and MORGAN, C. A. (1995). *Animal Nutrition*. 5th Edition, Pearson Education Limited, Edinburg Gate, Garlow Essex CM2O 2JE, United Kingdom.

NWAKOLOR, I. M. (2001). Crossing breeding of large white and Nsukka local pigs: genotype and age are related changes in haematological parameters. *Tropical Journal of Animal Science,* 4(2): 85 – 96.

PLUMB, D.C. (1999). *Veterinary Drug Handbook.* Iowa State University Press, Iowa, USA.

RANJHAN, S. K. (1993). *Animal Nutrition and Feeding Practices*. 4th Edition, Vikcas Publishing House, PVT Limited, New Delhi, India.

REITMAN, S. and FRANKEL, S. (1957). A colorimetric method for determination of serum glutamic oxaloacetic and glutamic pyruvic transaminase. *American Journal of Clinical Pathology,* 28: 56 – 62.

SAS (2008). *SAS/STAT User's Guide Version 8.* 3rd Edition, Statistical Analysis Systems (SAS) Institute Incorporated, Cary, North Carolina, USA.

SOBAMIWA, O. and LONGE, O. G. (1994). Utilization of cocoa-pod pericarp fractions in broiler chick diets. *Animal Feed Science and Technology*, 47(3-4): 237 – 244.

THRALL, M. A. and WEISER, M. G. (2002). Haematology. Pages 29 – 74. *In*: HENDRIX, C. M. (Ed.). *Laboratory Procedures for Veterinary Technicians*. 4th Edition, Mosby Incorporated, Missouri, USA.

WCF (2012). Cocoa Market Update. Compiled by the World Cocoa Foundation (WCF) from Published Reports and Resources. 7 pp. http://www.worldcocoafoundation.org/wp-content/uploads/Cocoa Market-Update-as of-3.20.2012.pdf