



## **Biotechnological Upgrade of High Fiber-Low Protein Industrial Plant By-products in Broiler Diets: Haematology and Serum Evaluation**

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### **Authors' contributions**

*This work was carried out in collaboration between both authors. Author AOF designed, supervised and edited the manuscript. Author DA was part of the research team that carried out the study on the Teaching & Research Farm of Ekiti State University. The both authors read and approved the final manuscript.*

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### **ABSTRACT**

**Aims:** To investigate some haematological parameters of broilers in which composite of bio-fermented industrial plant by-products was used as crude protein supplements.

**Study Design:** 288 birds were picked and randomized into 6 treatments in a randomized entirely designed experiment. The data collected were subjected to One Way Analysis of Variance (ANOVA) using Minitab computer model (Version 16).

**Place and Duration of Study:** Research study was carried out at the Teaching & Research Farm of Ekiti State University, Ado-Ekiti, a town in the Southwest Nigeria between June and September, 2016.

**Methodology:** A composite (PBMC) of the palm kernel meal, brewer dried grains and molasses were prepared and incorporated on dry matter basis as protein supplement at 15, 20, 25, 30 and 35% inclusion levels in the experimental diets.

**Results:** The haemoglobin concentration (HBC) value of birds on diet with zero PBMC had the

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lowest significant ( $P < 0.05$ ) value of  $6.58 \pm 0.01$  g/dl. Diet on 30% PBMC inclusion had the highest HBC value of  $9.06 \pm 0.06$  g/dl. The packed cell volume (PCV), red blood cell (RBC) and mean corpuscular volume (MCV) of birds on zero PBMC inclusion were consistently lower ( $P < 0.05$ ) than for other birds. The mean cell hemoglobin (MCH) were similar ( $P > 0.05$ ) for all experimental birds. The mean corpuscular hemoglobin concentration (MCHC) obtained for birds on zero PBMC inclusion level had the highest significant value of  $42.95 \pm 1.20$  g/dl but similar ( $P < 0.05$ ) to  $35.92 \pm 0.26$  g/dl for birds on 25% PBMC inclusion. The erythrocyte sedimentation rate (ESR) was lowest ( $P < 0.05$ ) in birds on 35% PBMC inclusion at  $5.15 \pm 0.07$  mm<sup>3</sup>/l and highest ( $P < 0.05$ ) in birds on 15% PBMC at  $5.90 \pm 0.14$  mm<sup>3</sup>/l. The highest ( $P < 0.05$ ) serum protein of  $34.91 \pm 0.14$  was obtained for birds on zero PBMC inclusion level. No significant differences ( $P > 0.05$ ) existed among the albumin-globulin ratio for all experimental birds. The investigated blood parameters indicated high degrees of similarity in values obtained for most haematological and serum indices in all experimental diets including the control diets.

**Conclusion:** PBMC inclusion levels at measured quantities not exceeding 35% did not have any deleterious effects on the response criteria measured in broilers and were all within normal reported ranges.

**Keywords:** Haematological parameters; total serum protein; health status.

## 1. INTRODUCTION

The advancement in the poultry industry is tremendous world over. The poultry industry is continuously advancing by improvement of the genetic potential of new broiler strains [1] to provide the high-quality with low-cost protein requirements of the human population worldwide. Genetic development for rapid growth together with intensive husbandry conditions has accelerated the outbreaks of avian diseases. Measurement of haematological parameters provides valuable information in this regards and routinely used in human's and animal's medicines, but unfortunately, due to lack of adequate information, blood profile have not been widely used in avian medicine [2,3]. Avian blood differs in cells' characteristics from their mammalian counterpart [4]. Several factors including diet contents [5,6] affect the blood profiles of healthy birds.

Brewer's dry grains (BDG) are the by-products of mashing process; which is one of the initial operations in the brewery in order to solubilize the malt and cereal grains to ensure adequate extraction of the wort (water with extracted matter) [7]. BDG is a readily available, high volume low-cost by-product of brewing and is a potentially valuable resource for industrial exploitation [8]. Thus, increased endogenous metabolism, as well as high proteolytic activity in BDG, affect its composition over a short period of time [9]. BDG is a relatively good source of protein and has been used in the feeding of pigs, sheep, and cattle [10]. The feeding capabilities of BDG is however, limited by its high crude fiber

content and low degradability of the crude fiber.

Palm kernel cake (PKC) as a case sample of agro-industrial by-products is an important end product of palm kernel oil (PKO) from the fruits of palm oil (*Elaeis guinensis*) which is readily available in many tropical environments among which are Nigeria, Malaysia, and Indonesia.

The utilization of fibrous feed materials by monogastric animals has been known to be a waste because, non-starch polysaccharides (NSPs) have the potency to confer some anti-nutritive activity. It was revealed that most fibrous feed components found in PKC are NSP and beta-mannan [11,12]. In this way, poultry cannot easily make use of such feed components for proper digestibility. However, the anti-nutritional factor associated with NSPs result to poor growth performance due to poor nutrient utilization by the animal. The biofermentation technology of processing both PKC and BDM for possible improved utilization in broilers will be reported in other studies by same author/s.

Available information on avian haematological studies is used in the present study as standards to investigate the blood profiles of broilers that were fed a composite of agro-industrial plant by-products.

This study was designed to investigate the haematological parameters of broiler birds in which bio-fermented composite of industrial plant by-products (palm kernel meal, PKM; brewer spent grains, BDG; and molasses) was used in

measured inclusion levels as crude protein supplements in broiler feeding.

## 2. MATERIALS AND METHODS

### 2.1 Experimental Site

The research study was carried out at the Teaching & Research Farm of Ekiti State University, Ado-Ekiti, a town in the Southwest Nigeria in the rain forest zone on latitude 7°40' North of the equator and longitude 5°15' East of the Greenwich Meridian with ambient temperature of 25-37°C; relative humidity, 70%; wind, SSW at 11mph (18 km/h); barometric pressure, 29.68' Hg (F) during the summer of year 2016.

### 2.2 Bio-fermentation Technology and Procedure for Ensiling

Palm kernel meals (PKM) were obtained from local communities (especially Ogotun-Ekiti) around Ado-Ekiti where palm oil is produced majorly by solvent extraction method. Brewers dried grains (BDG) were obtained from Nigerian Bottling Company, Ibadan through a reputable feed mill in Ado-Ekiti, Ekiti State. Mixtures of the PKM, BDG and molasses were prepared using a ratio of 50litres of water to 25 kg of PKM, 25 kg of BDG and 2.5litres of molasses. The mixture of PKM, BDG, molasses and water subsequently referred to as palm kernel, brewers spent grains and molasses composite (PBMC) was gently compressed into 120L plastic containers according to described method [13,14]. The compressing of the materials into containers was done manually at about 1 foot height interval until the containers were about  $\frac{3}{4}$  filled. The containers were carefully covered with thick nylon covering with sand used to fill the spaces left. There were further compressions and another thick nylon was spread across the rims of the containers before the containers were finally covered with their lids to ensure air-tightness. Containers containing the ensiled PBMC were opened on day 21 [13]. Samples were taken for laboratory analyses. The ensiled PBMC was later sun-dried to achieve a moisture content of 12%. Dried samples of the ensiled PBMC were then analysed for proximate acid composition before incorporation into feed formulation [15]

### 2.3 Experimental Ration Formulation

Samples of the BDG, PKM and PBMC were taken for proximate analyses after which they

were incorporated into the diets. The experimental diets were formulated as follows:

Diet 1 was the control diet without BDG and PKM mixture;

Diet 2 had PBMC at 10% inclusion level;

Diet 3 had PBMC at 15% inclusion level;

Diet 4 had PBMC at 20% inclusion level;

Diet 5 had PBMC at 25% inclusion level;

Diet 6 had PBMC at 30% inclusion level.

All diets were made isocaloric and isonitrogenous with ample supplementation of L-lysine and DL-methionine synthetic amino acids at required level [15]. Experimental diets were also taken into the laboratory for determination of proximate analyses [16].

### 2.4 Management of Experimental Birds

Two hundred and eighty eight were randomly picked after sexing on the 3rd day of the arrival of chicks for the experiment [17]. The chicks were brooded in a brooder house using electricity supplied constantly by 1KVA stand-by power generating plant at the Ekiti State University Teaching and Research Farms. A 5-day acclimatization period was observed before the commencement of the first phase (5-28days) of the experiment during which the broiler chicks were fed *ad libitum* on commercial chicks mash containing 23% crude protein (CP) before data collection. The chicks were managed on the floor for this phase of experiment. Appropriate veterinary routines were observed from day old. The experimental birds were randomly allocated into the 6 experimental treatments with 48 birds in each treatment while the each treatment was replicated 3 times. Each replicate had 16 birds as designed.

### 2.5 Data Collection from Haematological Parameters

At the end of the feeding trial, birds were randomly selected, weighed and sacrificed by severing the jugular vein and blood allowed to flow freely into labelled bottles one of which contained a speck of Ethylene-diamine-tetra-acetic acid (EDTA) while the other without EDTA was processed for serum. The blood samples were used for the determination of haematological parameters such as packed cell volume (PCV), red blood cell count (RBC), haemoglobin concentration (Hbc), total white blood cell count (WBC), differential count of white blood cell (leucocyte, neutrophil, monocyte,

basophil and eosinophil). The packed cell volume (PCV) was estimated by spinning about 75:1 of each blood sample in heparinized capillary tubes in an haematocrit micro centrifuge for 5 minutes while the total red blood cell (RBC) count was determined using normal saline as the diluting fluid.

The Haemoglobin concentration (Hbc) was estimated using cyanomethaemoglobin method as described [18], while the mean corpuscular haemoglobin concentration (MCHC), mean corpuscular volume (MCV), low density lipoprotein and globulin were calculated. Biochemical parameters such as total serum protein, albumin, cholesterol (total cholesterol, triglycerides, high density lipoprotein, low density lipoprotein) were estimated. Total serum protein was determined colorimetrically using the SIGMA assay kits. The albumin and the globulin were determined using the described method [19].

## 2.6 Statistical Analysis

The data collected for different parameters were subjected to analysis of variance (ANOVA) using statistical One Way Analysis of Variance of the Minitab computer model (Version 16) [20].

## 3. RESULTS AND DISCUSSION

The experimental diets for broilers at starter and finisher phases of production are presented in Tables 1 and 2.

The results obtained for haematology of broiler birds fed composite of fermented PKM, BDG and molasses is presented in Table 3.

The haemoglobin concentration value of birds on control diet (diet 1) had the lowest value of  $6.58 \pm 0.01$  g/dl and it was significantly different ( $P < 0.05$ ) from other treatments as compared to diet with 30% PBMC (diet 5) with the highest value of  $9.06 \pm 0.06$  g/dl. However, no significant differences ( $P > 0.05$ ) existed among birds on 25% PBMC diet (diet 4), 30% PBMC diet (diet 5) and 35% PBMC diet (diet 6) with the values of  $9.03 \pm 0.02$  g/dl,  $9.06 \pm 0.06$  g/dl and  $8.97 \pm 0.08$  g/dl, respectively.

The packed cell volume of birds on control diet (diet 1) had the lowest value of  $22.01 \pm 1.14\%$  and highest value of  $24.85 \pm 0.21\%$  was obtained from birds on 25% PBMC diet (diet 4). However, statistically, no significant differences ( $P > 0.05$ ) existed among all the treatments. The PCV range in the present study of 22.01-24.85 agreed

with the normal range of 24.9-45.2% obtained for Nigerian chickens [22]. This also agreed with a range of 17-23% in another study [23] where the effects of graded levels of PKC on hematological profiles of pullets were examined.

The red blood cell of birds on the control diet (diet 1), had the lowest value of  $1.5 \pm 0.02$  mm<sup>3</sup> and this was significantly different ( $P < 0.05$ ) from other treatments. However, the highest RBC value obtained for birds on 30% PBMC diet (diet 5) with the value of  $1.87 \pm 0.05$  mm<sup>3</sup> had no significant variation from other RBC values obtained for birds on 15%, 20%, 25%, and 35% PBMC diets at  $1.80 \pm 0.02$  mm<sup>3</sup>,  $1.81 \pm 0.02$  mm<sup>3</sup>,  $1.78 \pm 0.03$ , and  $1.86 \pm 0.08$ , respectively. Similar results of significance differences were obtained with broilers fed biodegraded brewer dried grains [24]. Reductions in packed cell volume and red blood cell value have been suggested to be indicative of low protein intake or mild anaemia [25]. Another study conducted to determine the effect of including expeller-extracted canola meal (EECM) in diets for broilers on blood serum concentration and other growth parameters showed no effect of EECM on the blood hemoglobin content [26].

Mean corpuscular volume (MCV) had the lowest values for birds on control diet and birds on 15% PBMC diet (diet 2) with values of  $0.11 \pm 0.01$   $\mu$ l each. The highest MCV value of  $0.13 \pm 0.01$   $\mu$ l was obtained for birds on 20%, and 35% PBMC diets. However, no significant differences ( $P > 0.05$ ) existed among birds placed on all the PBMC diet levels.

The mean cell hemoglobin (MCH) obtained for birds placed on different dietary inclusions of PBMC (0%, 15%, 20%, 25% 30%, and 35%) showed no significant differences ( $P > 0.05$ ) among the treatments. However, 20% PBMC diets had the lowest value of  $4.29 \pm 0.14$   $\mu$ g, and the highest value was obtained from birds placed on 25% PBMC diet with the value of  $4.45 \pm 0.07$   $\mu$ g. The present result differs from a previous report [22] who reported significant differences in the MCH of broilers fed biodegraded BDG.

The mean corpuscular hemoglobin concentration (MCHC) obtained for birds on the control diet had the highest value of  $42.95 \pm 1.20$  g/dl and showed a significant difference ( $P < 0.05$ ) from other birds on other diets. Birds on 25% PBMC inclusion level had the lowest value of  $35.92 \pm 0.26$  g/dl but statistically similar ( $P > 0.05$ ) to the other MCHC values except for the MCHC value obtained for

birds on the control diet without PBMC. The present result is also contrary to the result in a previous study [24] where a MCHC range of 33.06-33.33 (g/dl) with no significant difference was reported in broiler fed biodegraded BDG.

**Table 1. Proximate analysis of palm kernel meal (PKM), brewers dried grains (BDG) and palm kernel meal-brewer dried grain-molasses composite (PBMC)**

Proximate composition, %	PKM	BDG	Molasses (cane)	PBMC (unfermented)	Fermented PBMC
Dry matter	86.5±2.1	87.5±4.3	22.9	87.1 <sup>b</sup> ±2.1	89.5 <sup>a</sup> ±3.1
Crude protein	19.1±3.0	22.1±2.7	0.3	20.2 <sup>b</sup> ±4.5	3.1 <sup>a</sup> ±4.2
Crude fibre	14.4±1.7	12.3±3.2	0.0	13.5 <sup>a</sup> ±2.1	10.1 <sup>b</sup> ±3.3
Ether extract	7.3±1.8	7.6±2.1	0.0	7.4 <sup>a</sup> ±4.1	3.5 <sup>b</sup> ±3.1
Ash	11.1±2.1	4.7±3.3	1.1	8.2±2.5	8.5±2.0
Nitrogen free extract	48.1±2.7	53.3±2.1	74.8	49.8 <sup>b</sup> ±3.2	4.8 <sup>a</sup> ±3.4
Metabolisable energy, Kcal-kg	2719	2485	1958	2634	2789

Mean ± Standard deviation; Proximate analysis values obtained for fermented and unfermented PBMC were compared using T-Test of Minitab (Ver. 16) statistical model [20]. Means in the same row with the different superscript are significantly different at (P>0.05); (a, b, c) Means in the same row with the different superscript are significantly different at (P<0.05).

ME, metabolizable energy = (0.860+0.629(GE-0.78CF) [21]; Palm kernel meal, PKM; brewers dried grains, BDG; palm kernel meal-brewer dried grain-molasses composite, PBMC

**Table 2. Experimental diets (starter phase, 1-28 days)**

Ingredients	Diets and % inclusion levels of PBM composite)					
	Diet 1 0%	Diet 2 10%	Diet 3 15%	Diet 4 20%	Diet 5 25%	Diet 6 30%
Maize (11.0% CP)	50.40	47.85	47.00	45.50	44.50	43.05
Wheat offals	10.00	10.00	10.00	10.00	10.00	10.00
Soyabean meal	33.50	26.00	24.00	19.00	15.00	11.00
Fish meal	2.00	0.00	0.00	0.00	0.00	0.00
Palm oil	0.00	0.00	0.00	1.00	1.00	1.00
*PBMC	0.0	10.0	15.0	20.0	25.00	30.0
Bone meal	2.5	2.5	2.5	2.5	2.5	2.5
Oyster shell/limestone	0.5	0.5	0.5	0.5	0.5	0.5
NaCl	0.3	0.3	0.3	0.3	0.3	0.3
DL-methionine	0.1	0.1	0.1	0.1	0.1	0.1
L-Lysine	0.1	0.1	0.1	0.1	0.1	0.1
**Premix	0.5	0.5	0.5	0.5	0.5	0.5
Total	100.0	100.0	100.0	100.0	100.0	100.0
<b>Calculated composition</b>						
Crude protein, %	23.06	23.07	23.06	23.06	23.05	23.04
Crude fibre, %	4.54	5.64	5.82	6.02	7.85	9.24
Ether extract, %	7.21	7.01	7.03	9.21	9.43	9.32
L-lysine	0.18	0.17	0.17	0.17	0.16	0.16
DL-methionine	0.19	0.17	0.17	0.17	0.17	0.17
***ME, Kcal/Kg	2893.3	2889.7	2884.0	2885.8	2879.1	2886.8
<b>Proximate composition</b>						
Crude protein, %	22.86	22.90	23.01	23.04	23.10	23.10
Crude fibre, %	4.87	5.02	5.31	6.34	7.56	8.12
Ether extract, %	6.09	6.52	6.64	7.24	7.87	8.14

\*PBMC, Palm kernel meal+Brewers dried grains+Molasses ensiled composite;

\*\*Premix contained vitamins A(10,000,000iu); D(2,000,000iu); E(35,000iu);K(1,900mg); B12 (19mg); Riboflavin(7,000mg); Pyridoxine(3,800mg); Thiamine(2,200mg); D Panthotenic acid(11,000mg); Nicotinic acid(45,000mg); Folic acid(1,400mg); Biotin (113mg); and trace elements as Cu(8,000mg); Mn(64,000mg); Zn(40,000mg); Fe(32,000mg); Se(160mg); I<sub>2</sub>(800mg); and other items as Co(400mg); Choline(475,000mg); Methionine(50,000mg); BHT(5,000mg) and Spiramycin(5,000mg) per 2.5kg; CP:Crude Protein, ME:Metabolized Energy; \*\*\*ME, metabolizable energy = (0.860+0.629(GE-0.78CF) [21]

**Table 3. Experimental diets (finisher phase, 29-58 days)**

Ingredients	Diets and % inclusion levels of PBM composite					
	Diet 1 0%	Diet 2 15%	Diet 3 20%	Diet 4 25%	Diet 5 30%	Diet 6 35%
Maize (11.0% CP)	55.40	42.85	42.00	40.50	39.50	39.05
Wheat offals	10.00	10.0	10.0	10.0	10.0	10.0
Soyabean meal	28.50	31.00	19.00	14.00	10.00	10.00
Fish meal	2.00	0.0	0.0	0.0	0.00	0.0
Palm oil	0.00	0.00	0.00	1.00	1.00	1.00
*PBM	0.00	15.0	20.0	25.0	30.00	35.0
Bone meal	2.5	2.5	2.5	2.5	2.5	2.5
Oyster shell/limestone	0.5	0.5	0.5	0.5	0.5	0.5
NaCl	0.3	0.3	0.3	0.3	0.3	0.3
DL-methionine	0.1	0.1	0.1	0.1	0.1	0.1
L-lysine	0.1	0.1	0.1	0.1	0.1	0.1
**Premix	0.5	0.5	0.5	0.5	0.5	0.5
Total	100.0	100.0	100.0	100.0	100.0	100.0
<b>Calculated composition</b>						
Crude protein, %	20.13	20.07	20.01	20.02	19.65	19.60
Crude fibre, %	5.14	5.78	6.12	7.21	8.91	9.89
Ether extract, %	7.34	7.41	7.53	9.34	9.43	9.43
L-lysine	0.18	0.17	0.17	0.17	0.16	0.16
DL-methionine	0.19	0.17	0.17	0.17	0.17	0.17
***ME, Kcal/Kg	3101.4	3210.3	3120.3	3103.8	3134.4	3103.8
<b>Proximate composition</b>						
Crude protein, %	20.06	20.10	19.87	20.01	19.89	19.90
Crude fibre, %	4.90	5.23	5.72	6.31	7.21	7.86
Ether extract, %	7.09	7.52	7.14	7.35	7.35	7.89

\*PBM, Palm kernel meal+Brewers dried grains+Molasses ensiled composite; \*\*Premix contained vitamins A(10,000,000iu); D(2,000,000iu); E(35,000iu);K(1,900mg); B12 (19mg); Riboflavin(7,000mg); Pyridoxine(3,800mg); Thiamine(2,200mg); D Panthotenic acid(11,000mg); Nicotinic acid(45,000mg); Folic acid(1,400mg); Biotin (113mg); and trace elements as Cu(8,000mg); Mn(64,000mg); Zn(40,000mg); Fe(32,000mg); Se(160mg); I<sub>2</sub>(800mg); and other items as Co(400mg); Choline(475,000mg); Methionine(50,000mg); BHT(5,000mg) and Spiramycin(5,000mg) per 2.5kg; CP:Crude Protein, ME:Metabolized Energy;

\*\*\*ME, metabolizable energy = (0.860+0.629(GE-0.78CF) [21]

The erythrocyte sedimentation rate (ESR) was lowest in birds on the diet with 35% PBM inclusion level with the value of 5.15±0.07 mm<sup>3</sup>/l and highest in birds on 15% PBM diet with the value of 5.90±0.14 mm<sup>3</sup>/l. However, no significant differences (P>0.05) existed among the birds on all the diets. This present result was similar to a previous study where no significant differences (P>0.05) were reported for ESR values of birds fed fermented cassava pulp [27].

Except for lymphocytes with slightly significant variations (P>0.05) ranging from the lowest value of 59.0±0.82% in birds on zero PBM inclusion to 62.25±0.50 in birds on both diets with 30% and 35% PBM inclusion levels, other white blood cell parameters (neutrophils, monocytes,

basophils and eosinophils) all showed similar (P>0.05) values across the dietary treatments of different PBM inclusion levels.

The results obtained for serum analysis of broiler birds fed composite of fermented PKM, BDG and molasses is presented in Table 4.

The total serum protein (TSP) obtained for birds on control diet (diet 1) had the highest value of 34.91±0.14 with significant differences (P<0.05) existing between it and TSP values obtained for birds on other diets. The lowest value was obtained for birds on diet with 20% PBM inclusion level (diet 3) with the value of 31.25±0.35. However, statistical similarity (P>0.05) was obtained for birds on other diets (diets 2, 3, 4, 5, and 6).

**Table 4. Haematology of broiler birds fed composite of fermented PKM, BDG and molasses**

Parameters	Diets and % inclusion levels of PBM composite					
	Diet 1	Diet 2	Diet 3	Diet 4	Diet 5	Diet 6
	Inclusion levels of palm kernel meal, brewer dried grains and molasses composite, PBMC					
	0%	15%	20%	25%	30%	35%
Hbc (g/dl)	6.58 <sup>d</sup> ±0.01	8.19 <sup>c</sup> ±0.03	8.55 <sup>b</sup> ±0.50	9.03 <sup>a</sup> ±0.02	9.06 <sup>a</sup> ±0.06	8.97 <sup>a</sup> ±0.08
PCV, %	22.01 <sup>a</sup> ±1.14	22.55 <sup>a</sup> ±0.78	23.95 <sup>a</sup> ±0.35	24.85 <sup>a</sup> ±0.21	24.77 <sup>a</sup> ±0.06	24.45 <sup>a</sup> ±0.93
RBC, 10 <sup>6</sup> /mm <sup>3</sup>	1.5 <sup>b</sup> ±0.02	1.80 <sup>a</sup> ±0.02	1.81 <sup>a</sup> ±0.02	1.78 <sup>a</sup> ±0.03	1.87 <sup>a</sup> ±0.05	1.5 <sup>b</sup> ±0.02
ESR, mm <sup>3</sup> /l	5.60 <sup>a</sup> ±0.57	5.90 <sup>a</sup> ±0.14	5.3 <sup>a</sup> ±0.14	5.20 <sup>a</sup> ±0.28	5.35 <sup>a</sup> ±0.35	5.15 <sup>a</sup> ±0.07
MCV ×10 <sup>-6</sup> (ul)	0.11 <sup>a</sup> ±0.01	0.11 <sup>a</sup> ±0.01	0.13 <sup>a</sup> ±0.01	0.12 <sup>a</sup> ±0.01	0.13 <sup>a</sup> ±0.01	0.12 <sup>a</sup> ±0.01
MCH ×10 <sup>-6</sup> (ug)	4.40 <sup>a</sup> ±0.02	4.40 <sup>a</sup> ±0.04	4.29 <sup>a</sup> ±0.14	4.45 <sup>a</sup> ±0.07	4.44 <sup>a</sup> ±0.10 <sup>a</sup>	4.39 <sup>a</sup> ±0.01
MCHC	42.95 <sup>a</sup> ±1.20	38.65 <sup>b</sup> ±0.73	35.93 <sup>b</sup> ±1.08	35.92 <sup>b</sup> ±0.26	37.00 <sup>b</sup> ±0.28	36.45 <sup>b</sup> ±1.06
Lymphocytes	59.00±0.82 <sup>c</sup>	60.50±0.58 <sup>bc</sup>	61.00±0.82 <sup>ab</sup>	62.00±0.82 <sup>ab</sup>	62.25±0.50 <sup>a</sup>	62.25±0.50 <sup>a</sup>
Neutrophils	26.25±2.99	23.50±2.65	24.25±3.10	22.75±1.71	22.50±2.38	22.25±0.96
Monocytes	11.75±2.50	13.00±2.58	11.50±2.52	12.50±2.08	12.00±2.45	12.75±0.96
Basophils	2.50±0.58	2.25±0.50	2.25±0.50	2.00±0.00	2.25±0.50	2.25±0.50
Eosinophils	1.00±0.00	0.75±0.50	1.00±0.00	0.75±0.50	1.00±0.00	1.00±0.00

Mean ± Standard deviation; (a, b, c) Means in the same row with the different superscript are significantly different at (P<0.05). PCV=Packed Cell Volume, Hbc=Haemoglobin concentration, RBC=Red blood cell, TWBC=Total white blood cell, MCH=Mean cell haemoglobin, MCHC=Mean cell haemoglobin concentration, MCV=Mean cell volume

**Table 5. Serum biochemical parameters of broiler birds fed composite of fermented PKM, BDG and molasses**

	Diets and % inclusion levels of PBM composite					
	Diet 1	Diet 2	Diet 3	Diet 4	Diet 5	Diet 6
	Inclusion levels of palm kernel meal, brewer dried grains and molasses composite, PBMC					
	0%	15%	20%	25%	30%	35%
TSP, g/dl	34.91 <sup>a</sup> ±0.14	32.55 <sup>b</sup> ±0.64	31.25 <sup>b</sup> ±0.35	32.00 <sup>b</sup> ±0.28	31.40 ±0.57	31.90 <sup>b</sup> ±0.14
Albumin, g/dl	18.54 <sup>a</sup> ±0.67	17.32 <sup>b</sup> ±0.09	16.96 <sup>b</sup> ±0.06	17.07 <sup>b</sup> ±0.04	17.06 <sup>b</sup> ±0.06	17.20 <sup>b</sup> ±0.16
Globulin, g/dl	16.38 <sup>a</sup> ±0.53	15.23 <sup>ab</sup> ±0.55	14.30 <sup>b</sup> ±0.30	14.94 <sup>ab</sup> ±0.25	14.3 <sup>b</sup> ±0.50	14.7 <sup>b</sup> ±0.02
Alb/Glb, g/dl	1.14±0.08	1.14±0.04	1.19±0.02	1.14±0.01	1.19±0.04	1.17±0.01

TSP, total serum protein; Alb, albumin; Glb, globulin Mean ± Standard deviation; (a, b, c) Means in the same row with the different superscript are significantly different at (P<0.05)

The albumin in the serum of birds on the control diet had the highest value of  $18.54 \pm 0.67$  and this was significantly higher ( $P < 0.05$ ) than all other albumin values. However, all other albumin values for birds on diets 2, 3, 4, 5 and 6 had similar ( $P > 0.05$ ) values. This present result agreed with the study where birds were fed fermented cassava pulp and significant differences were reported [27] but disagreed with another previous study [24] who reported no significant difference ( $P > 0.05$ ) in the albumin of broiler fed biodegraded BDG.

The globulin in the serum of birds on control diet had the highest value of  $16.38 \pm 0.53$  and the lowest value was obtained from birds on diet with 20% PBMC inclusion level (diet 3) with the value of  $14.30 \pm 0.30$ . However, significant differences ( $P < 0.05$ ) existed among birds placed on different diets. The highest globulin value of  $16.38 \pm 0.53$ g/dl was obtained for birds on zero PBMC diet but similar ( $P > 0.05$ ) to  $15.23 \pm 0.55$ g/dl and  $14.94 \pm 0.25$ g/dl obtained for birds on 15% and 25% PBMC inclusion levels, respectively. The lowest globulin value of  $14.3$ g/dl was obtained for birds on 20% and 30% PBMC, respectively. This is in line with previous study [24] where significant differences ( $P < 0.05$ ) in globulin levels of broiler fed biodegraded BDG were reported. However, another study had a different report [27] when broilers were fed with fermented cassava pulp showed no significant differences ( $P > 0.05$ ) in the globulin levels.

Albumin globulin ratio for birds on 25% PBMC diet had the lowest value of  $1.14 \pm 0.01$  and highest value was obtained for birds on 30% PBMC diet with the value of  $1.19 \pm 0.04$ . Despite the numerical differences, no significant differences ( $P > 0.05$ ) existed among the birds on all the diets.

The range of albumin globulin ratio values of  $1.14 \pm 0.01$  to  $1.19 \pm 0.04$  differed from the reported values [24] with significant differences ( $P < 0.05$ ) in the albumin globulin ratio of broiler fed fermented cassava pulp.

The measure of the serum globulin content and A/G ratio (either low or high), may show a better disease resistance and immune response of chicken [28].

The haematological values of the experimental birds indicated that most investigated parameters were within normal range [29,30].

#### 4. CONCLUSION

The investigated blood parameters indicated high degrees of similarity in values obtained for most haematological and serum indices in all experimental diets including the control diets without PBMC inclusion. The introduction of PBMC at measured quantities into broiler diets did not have any deleterious effects on the response criteria measured. The haematological values of the experimental birds indicated that most investigated parameters were within normal range. There is no predisposition to acute general infection, malignant tumor, malformation or diseased condition even at the highest inclusion level of PBMC 35% in broiler finisher diets.

#### ETHICAL DISCLAIMER

As per international standard or university standard written ethical approval has been collected and preserved by the author(s).

#### 5. RECOMMENDATION

Barring other nutritional factors which are reported in related studies supervised by the same author, it is safe and possible to incorporate palm kernel meal, brewer dried grains and molasses composite (PBMC) within 35% inclusion levels into broiler feed for effective utilization without any adverse health implication.

#### COMPETING INTERESTS

Authors have declared that no competing interests exist.

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