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Evaluating the Potentials of Carica papaya seed as phytobiotic to improve feed efficiency, growth performance and serum biochemical prameters in broiler chickens

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Abstract: *Sixty day old chickens were randomly distributed into three treatments and fed diets mixed with 0, 0.5 and 1% papaya seed powder for the six weeks' starter period. Feed consumption and growth performance were monitored weekly. Lipid peroxidation was also assayed in the serum of the chicks from all treatments. At the end of the experiment, results showed that feed efficiency and growth performance of broilers were not significantly (p>0.05) influenced by dietary treatments. The haematology parameters such as red blood cell, haemoglobin, were significantly (p<0.05) influenced by dietary inclusion of papaya seed powder. There were no significant (p>0.05) influence of papaya seed powder on serum biochemical parameters. Furthermore, lipid peroxidation decreased significantly (p<0.05) in treatments with papaya seed powder when compared to the control. It may be concluded that feeding broiler chickens with diets mixed with papaya seed powder significantly and positively reduced serum lipid peroxidation profile without negatively affecting feed efficiency, growth performance and serum biochemical parameters.*

Key words: *Phytobiotic, Pawpaw seed powder, broiler, antioxidant, lipid peroxidation*

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I. Introduction

Poultry farming refers to the process of rearing domestic birds such as chickens, ducks turkeys, geese, etc for the purpose of farming meat or eggs for food[1]. In Africa, agriculture and agro-industries account for over 30% of national incomes on average, as well as for the bulk of export revenues[2]. Nearly three-quarters of the African population depend on agriculture to secure their livelihoods[3 4]. As a result of high population growth in Africa and the growing income, the demand for eggs and poultry meat has significantly increased in recent years across large parts of the African continent[5]. Based on estimates by the United States Agency for International Development, this trend is likely to continue over the next few years[2]. Therefore, the consumption of poultry and eggs is projected to increase by about 200% between 2010 and 2020 in some countries in sub-Saharan Africa, including Nigeria[6 7].

To increase production, use of feed additives in animal feeding began in the 1940s, to improve the organoleptic characteristics of raw materials, fodders, and/or animal products, and to prevent diseases[8 9]. Additives are also used to improve production efficiency by decreasing the mortality rate and stimulating weight gain[10 11 12]. Different categories are common depending on their properties and functions[13].

Unfortunately, the risk posed by antibiotic growth promoters (AGPs) to create cross-resistance to antibiotics used in human medicine and their presence in animal products, has led to their use to drop significantly, this is as they are banned in some cases in the formulation of fodders and for use in general animal husbandry[14]. Some researchers however, suggested that the ban on these substances has caused increase in the incidences of bacterial infections (i.e., diarrhea, coccidiosis and intestinal necrosis)[14 11]. The prohibitions on AGPs resulted in economic impact on the livestock industry because it led to increased production costs. The American livestock industry demonstrated that the use of AGP in poultry production was associated with losses for producers[12 16].

The restrictions on AGPs has stimulated interest in medicinal plants, which was revived in recent times because of their efficacy in providing cost effective therapy to several diseases because of secondary metabolites abundant in plants. Interestingly, Papaya (Carica papaya) is rich in polyphenols which makes them versatile tools for the treatment of ailments in folklore medicine[17]. Papaya is a common human fruit; available throughout the year in the tropics. It is referred to as the "medicine tree" or "melon of health", and the major active ingredients recorded include; carpine, chymopapain and papain, a bactericidal aglycone of glucotropaeolin, benzyl isothiocyanate, a glycoside sinigrin, the enzyme myrosin, and carpasemine[18]. This present study was designed to investigate the effects of dried Carica papaya seed powder on growth performance, feed efficiency, antioxidant enzyme activity, and serum biochemical parameters in broiler chickens.

II. Materials and Method

Sixty day-old broiler starter birds were purchased from a commercial supplier in Minna, Niger state, and transported to the research facility in ventilated paper cages. Day old broiler chicks were exposed to cross-ventilation during the period of the study. Chicks were reared in cages of dimension 1.5 m x 1.5 m x 2.0 m; length x breadth x height, that were previously cleaned and disinfected. Chicks were acclimated for 5 days. During the acclimation, the chicks were fed commercial starter feed (23.62% Crude Protein, 14.7% Crude fat) and water was supplied ad libitum under strict biosecurity control according to previously published protocol[19].

Study location: The feeding trial phase of this research study was carried out at the aquatic animal research facility of biochemistry department Ibrahim Badamasi Babangida University, Lapai, Niger State.

Study design: Sixty day old chickens were randomly distributed into three treatments and fed commercial starter feed (Chikun, Olam poultry feed mill, Kaduna, Nigeria) mixed with 0, 0.5 and 1% PSP for the six weeks' starter period. Pre-weighed feed were provided to all treatments every morning to monitor feed intake while drinking water were provided. Feed and water were offered to the birds in all the different treatments ad-libitum. All birds were reared without vaccination. Chicks were reared under approximately natural photoperiod of 12/12 hours of light/dark cycles for period of six weeks and cages were cleaned at the end of every week.

Study duration: The experiment lasted for a period of 6 weeks of 7 days each.

Sample size: Sixty day-old broiler chicks'

Procedure methodology

Proximate composition analysis of experimental feeds and papaya seeds powder

Moisture content, crude fat, crude fiber, and crude protein (Microkjeldahl N x 6.25) were all determined following standard Methods of the Association of Analytical Chemists[20].

Vitamin analysis

Vitamin analyses including vitamin C (ascorbic acid) and beta-carotene were determined in sample (papaya seed powder) according to the method described by 2, 6-dichlorophenol indophenols method of Eleri and Hughes[21] whereas; Tocopherol was estimated in the sample by the Emmerie-Engel reaction as reported by Rosenberg[22].

Calculations for feed efficiency and growth performance parameters

Various parameters were calculated by applying the appropriate formulae where necessary, using the following:

Chicks survival (%) = (total chicks' survival/ total chicks stock) x 100

Weight gain %(WG %) = $[(W_f - W_i)/W_i] \times 100$

Specific growth rate (SGR %) = $[(\ln W_f - \ln W_i)/T] \times 100$

Feed intake (FI) = total feed intake/number of live chicks

Feed conversion ratio (FCR) = wet weight gain (g)/feed intake (g)

Where W_f refers to the mean final weight, W_i is the mean initial weight and T is the feeding trial period in days.

Determination of haematological parameters

Haematological components including red blood cells (RBC), haemoglobin (HGB), Mean cell volume (MCV), hematocrit (HCT%), Platelet number (PLT), white blood cells (WBC), lyphocytes (LYM), Mean cell haemoglobin, (MCH), Mean cell haemoglobin concentration (MCHC), Mean platelet volume (MPV), were determined using the automated haematologic analyzer (ABACUS 3CT, Diatron, USA), employing the methods described by Dacie and Lewis[23].

Determination of Serum biochemical analysis

Chicks serum biochemical analysis such as AST, ALT, ALP, bilirubin, albumin, creatinine, proteins, potassium, chloride, sodium and urea were determined spectrophotometrically following the manufacturer's instructions of AGAPPE diagnostics, Cham, Switzerland while Lipid peroxidation was assayed according to the method described by Health and Parker[24].

Determination of activities of antioxidant enzymes

Antioxidant enzymes including superoxide dismutase (SOD) and catalase (CAT) were determined spectrophotometrically from chicks' serum according to the method described by Yusuf, where lipid peroxidation was assayed according to the method described by Health and Parker[24].

Statistical analysis

Data obtained were subjected to one-way analysis of variance (ANOVA) and the means were compared using Turkeys test. Statistical significance was set at $P < 0.05$. Statistical analyses were performed using SPSS software Version 20.0. Data for all determinations are presented as mean ± SEM of three replicates.

III. Results

No chickens died during the experiment, as 100 % of the chickens from all treatments survived. Results for proximate composition analysis of PSP and all dietary treatments are shown in Table 1. From the Table, it is observed that PSP is a good source of fat, protein and fibre. The moisture content of PSP was low. The result also showed moderate value of ash content that suggests papaya would provide essentials minerals. Furthermore, PSP also contains antioxidant enzymes such as vitamin C, E and β-carotene. Inclusion of PSP up to 1 % did not significantly ($P > 0.05$) modify the nutrient composition of all dietary treatments compared to the control.

Table 1: Proximate composition of experimental feeds and Carica papaya seed powder

		Treatments		
Parameters	PSP	0 % PSP	0.50 % PSP	1 % PSP
MC %	6.81±0.00	8.44±0.01[a]	7.56±0.01[a]	7.66±0.01[a]
CF %	30.80±0.03	4.90±0.01[a]	5.90±0.01[a]	6.50±0.01[a]
ASH %	2.09±0.00	4.30±0.01[a]	5.95±0.01[a]	7.10±0.01[a]
FAT %	28.19±0.00	14.70±0.01[a]	12.0±0.01[a]	13.67±0.01[a]
CP %	26.25±7.99	23.62±0.01[a]	27.12±0.01[a]	32.37±0.01[a]
CHO %	5.86±0.01	44.04±0.01[a]	34.97±0.01[a]	39.20±0.01[a]
Vit C (mg/100g	0.85±0.02			
Vit E (µg/100g	2.00±0.33			
β-carotene (mg/100g)	0.40±0.03			

Data are expressed as mean ± SEM (n = 20).
Mean ± SEM followed by different superscript letter within a row are significantly different ($P < 0.05$).
MC, Moisture content; CF, Crude fibre; CP, Crude protein; CHO, Carbohydrate (Nitrogen free extract).

Table 2 showed the results for feed efficiency and growth performance of broiler chickens fed graded level of PSP for starter period. The data indicated that weight gain increased with increasing level of PSP in the diet from 0.5% to 1.0 %. Generally, better feed conversion ratio values were obtained in all treatments, but getting poor with increase of dietary PSP up to 1 %. Chicks fed diet containing 0.5 % PSP had better feed conversion ratio compared to the control. Feed intake was significantly ($P < 0.05$) the highest in 1% PSP.

Table 2: Feed efficiency and growth performance of broiler chickens fed graded level of C. papaya seed powder for 6 weeks starter period.

	Dietary treatments		
Parameters	0 % PSP	0.50 % PSP	1 % PSP
Growth performance			
Mean initial weight (g)	69.20±2.44[a]	66.55±1.92[a]	68.25±1.48[a]
Mean final weight (g)	2001.7±57.81[a]	2079.75±60.46[a]	2175.67±82.79[b]
Specific growth rate (%)	8.00±0.24	8.19±0.04	7.88±0.05
Weight gain (g)	1932.50±0.12[a]	2013.25±0.19[a]	2085.42±0.16[a]
Av. Daily weight gain (g)	46.00±0.31	47.93±0.04	49.64±0.21
Feed efficiency			
Feed intake (g)	3498.75±0.25[a]	3462.75±0.25[a]	4084.28±0.09[b]
Feed conversion ratio	1.81±0.00	1.72±0.01	1.96±0.02

Data are expressed as mean ± SEM (n = 20).
Mean ± SEM followed by different letter within a row are significantly different ($P < 0.05$).

Haematology parameters measured in broiler chickens fed graded level of PSP diets are as shown in

Table 3. From the Table, the haematology parameters such as MCV, HCT% and PLT did not significantly ($P > 0.05$) differ across all the treatments. RBC and HGB values recorded were higher ($P < 0.05$) in 0 % free PSP diet group compared to 0.5 % PSP supplemented group and it became lower with the increased in PSP up to 1 % PSP. Chicks fed 0.5 % PSP had higher but not significant ($P > 0.05$) MCV value when compared to control. Furthermore, chicks fed diet containing 1 % PSP recorded the least MCV value. HCT which is the percent of whole blood made up of red blood cell showed a dose dependent effect of PSP supplementation.

Table 3: Haematology parameters of broiler chickens fed graded level of c. papaya seed powder for 6 weeks starter period.

	Treatments		
Parameters	0 % PSP	0.5 % PSP	1 % PSP
RBC ($\times 10^6 mm^{-3}$)	8.49±2.58[a]	6.37±0.40[b]	3.83±0.63[c]
HGB (g/dl)	14.63±0.20[a]	13.93±0.22[ab]	11.87±3.37[b]
MCV (m^3)	126.67±0.33[a]	127.67±1.33[a]	125.67±1.20[a]
HCT (%)	28.35±0.21[a]	27.61±0.73[a]	23.51±6.89[a]
PLT (mcL)	3.00±0.57[a]	2.33±0.88[a]	3.00±2.52[a]

Data are expressed as mean ± SEM (n = 3).
Mean ± SEM followed by different letter within a row are significantly different ($P < 0.05$).
RBC, Red blood cell number; HGB, Haemoglobin; MCV, Mean cell volume; HCT, Haematocrit; PLT, Platelet number.

The result of blood parameters of broiler chickens fed graded level of PSP diets are as shown in Table 4. Blood parameters such as WBC, LYM, MCH, MCHC and MPV did not significantly ($P > 0.05$) differ across all the treatments. The result of MCH, which is the average amount of HGB in each RBC measured, followed the same pattern as MCV.

Table 4: Blood parameters of broiler chickens fed graded level of C. papaya seed powder for 6 weeks starter period.

	Treatments		
Parameters	0% PSP	0.5 % PSP	1% PSP
WBC ($\times 10^6 mm^{-3}$)	89.20±0.91[a]	74.34±12.53[a]	42.02±12.08[a]
LYM (%)	62.37±5.62[a]	54.07±12.94[a]	30.75±9.61[a]
MCH (pg)	65.20±0.50[a]	64.53±0.92[a]	63.87±1.30[a]
MCHC (g/dL)	51.50±0.45[a]	50.60±0.80[a]	50.83±0.90[a]
MPV (fL)	9.87±1.78[a]	10.40±1.16[a]	5.80±2.99[a]

Data are expressed as mean ± SEM (n = 3).
Mean ± SEM followed by different letter within a row are significantly different ($P < 0.05$).

WBC, White blood cell number; LYM, Lymphocytes; MCH, Mean cell haemoglobin; MCHC, Mean cell haemoglobin concentration; MPV, Mean platelet volume.

Results for hepatic and renal function biomarkers of broiler chickens fed graded level of PSP for 6 weeks starter period are shown in Table 5. The result showed no significant difference ($P > 0.05$) across all the treatments, of the biomarkers monitored. Although, values of AST and ALT obtained in broiler chicks fed PSP containing diet were lower when compared to the control diet (0 % PSP) which, however, still falls within the normal range in chickens which ranged from 1-37 µL[25].

Table 5: Hepatic and Renal function biomarker of broiler chickens fed graded level of C. papaya seed powder for 6 weeks starter period.

	Treatments		
Parameters	0% PSP	0.50% PSP	1% PSP
AST (µL)	16.80±1.25^{a}	16.20±0.46^{a}	15.17±1.89^{a}
ALT (µL)	18.90±0.76^{a}	17.93±2.72^{a}	17.97±0.59^{a}
ALP (µL)	25.97±1.86^{a}	27.23±1.88^{a}	28.87±0.82^{a}
Bil-total (mg/dl)	1.93±0.14^{a}	1.87±0.23^{a}	1.47±0.18^{a}
Bil-direct (mg/dl)	1.43±0.07^{a}	1.67±0.23^{a}	1.43±0.03^{a}
Albumin (µl)	1.63±0.07^{a}	1.43±0.07^{a}	1.63±0.03^{a}

Data are expressed as mean ± SEM (n = 3).
Mean ± SEM followed by different letter within a row are significantly different (P <0.05).
AST, Aspertate aminotransferase; ALT, Alanin aminotranferase; ALP, Alkline phosphate.

The result of serum metabolic parameters of broiler chickens fed graded level of PSP diets are as shown in Table 6. The result of serum metabolic parameters; total protein, creatinine, urea, potassium, chloride and sodium did not significantly (P > 0.05) differ across all the treatments. Creatinine values of broiler chickens fed PSP containing diet were numerically not significant (P > 0.05) different compared to the control.

Table 6: Serum metabolic parameters of broiler chickens fed graded level of C. papaya seed powder for 6 weeks starter period.

	Treatments		
Parameters	0 % PSP	0.5 % PSP	1 % PSP
Total protein (g/dl)	1.70±0.06^{a}	1.67±0.09^{a}	2.00±0.15^{a}
Creatinine (mg/dl)	1.40±0.11^{a}	1.30±0.06^{a}	1.27±0.03^{a}
Urea (mg/dl)	46.50±0.26^{a}	46.57±0.26^{a}	47.63±0.43^{a}
Potassium (mmol/l)	3.43±0.22^{a}	3.90±0.21^{a}	3.47±0.13^{a}
Chloride (meq/l)	53.23±1.57^{a}	61.33±3.56^{a}	95.43±17.09^{a}
Sodium (meq/l)	114.17±1.82^{a}	148.57±4.37^{a}	127.07±5.47^{a}

Data are expressed as mean ± SEM (n = 3).
Mean ± SEM followed by different letter within a row are significantly different (P <0.05).

Serum antioxidant enzymes (SOD and catalase) activities and LPO data for broiler chickens fed graded level of PSP diets are as shown in Table 7. The activities of superoxide dismutase and catalase as antioxidant enzymes were significantly (P<0.05) elevated in treatment with PSP when compared to the control (0 % PSP). The result of antioxidant enzyme activity (SOD and CAT) showed a significant (P<0.05) increase in 1 % PSP compared with the control, but 0.5 % PSP had no significant increase compared with the control. However, lipid peroxidation profile decreased significantly (P < 0.05) in treatment with PSP when compared to the control, and peroxidation decreased with more PSP in the diet.

Table 7: Serum antioxidant enzyme activity of broiler chickens fed graded level of C. papaya seed powder for 6 weeks starter period.

	Treatments		
Parameters	0 % PSP	0.5 % PSP	1 % PSP
SOD (μ/l)	2.60±0.23^{a}	3.03±0.03^{ab}	4.87±0.9^{b}
CAT (μ/l)	2.66±0.41^{a}	3.48±0.08^{ab}	4.14±0.08^{b}
LPO (nmol/ml)	5.98±1.33^{a}	2.86±0.04^{b}	1.77±0.21^{b}

Data are expressed as mean ± SEM (n = 3).
Mean ± SEM followed by different letter within a row are significantly different (P<0.05).
SOD, Superoxide dismutase; CAT, Catalase; LPO, Lipid peroxidation assay.

IV. Discussion

No chickens died during the experiment, as all (100 %) of the chickens from all treatments survived. The survival of all chickens from all treatment is an indication that PSP may not contain any toxic substances. No vaccines or AGPs were used in the study.

The crude protein content for PSP was noted to be high indicating that it is a very good source of protein (Table 1). Protein is an essential component of diet needed for the survival of both animal and human of which basic function is to supply adequate amount required[26]. Protein represent key nutrient for muscle and bone health, and thereby function in the prevention of osteoporosis in chickens[27]. The protein content of PSP noted in the present study was similar to that reported by Makanjuola[28] and Maisarah, et al.[29].

PSP was also noticed to be a good source of fiber. According to Eromosele[30] fibre helps in the maintenance of animal health and has been known to reduce cholesterol level in the body. A high fibre food expands the internal wall of the colon, causing the passage of waste, thus making it an effective anti-constipation. Fibre also reduces the risk of various cancers, bowel diseases and improves general health and well-being of animals. The crude fibre content of PSP recorded in the present study was similar to that reported by Adebisi, and Olagunju[31].

Fat content of PSP was recorded to be high indicating that PSP is a good source of oil which can serve as energy source. According to Wood, et al.[32] dietary intake of fat (unsaturated fatty acids) reduces the risk of cardiovascular disease and possibly the incidence of some cancers, asthma and diabetes in chickens. The fat content of PSP in the present study was similar to that reported by Makanjuola[28].

The moisture content of PSP was low. Low moisture contents generally are an indication of high shelf life especially for foods that are properly packaged against external condition[28]. Similar results were earlier reported by Chan-Prove et al.[33].

Furthermore, PSP also contains antioxidant enzymes such as vitamin C, E and β-carotene. Vitamin C content of PSP in this study was recorded to be low compared to previous literature[29]. Souza et al.[34] reported higher ascorbic acid content which was varied in two different species of papaya. Papaya is a source of vitamin C with amounts varying between the maturation stages [35 36]. Variation in vitamin C content was also reported among papaya varieties[37]. Vitamin C is an important antioxidant in aqueous phase, capable of scavenging oxygen derived free radicals[38]. Wall[37] suggested that papayas supply good quality of vitamins C and A. Papaya ranks first among 13-17 fresh fruits for vitamin C content[39].

PSP was also noted to contain Vitamin E in a moderate quantity. Vitamin E is one of the most important lipid-soluble primary defense antioxidants[40 41]. Vitamin E scavenges peroxyl radical intermediates in lipid peroxidation and responsible for protecting Poly Unsaturated Fatty Acid (PUFA) present in cell membrane and low density lipoprotein (LDL), against lipid peroxidation[42]. However Rojas and Campos[43] reported Vitamin E content for ripe papaya in moderate quantity and this was quite similar to that in the present study.

From the result of this study, beta-carotene was also noticed to be present in PSP. Beta-carotene has antioxidant properties that can help neutralize free radicals (reactive oxygen molecules) that potentially damage lipids in cell membranes and genetic material. This may lead to the development of cardiovascular disease and cancer[44]. Mean beta carotene content observed in this study was higher than previously reported by Souza et al.[34]. The differences in the beta carotene content observed could be attributed to methods of analysis that have been shown to contribute to variations in reported beta-carotene contents[45].

Inclusion of PSP up to 1 % did not significantly alter nutrient composition of all dietary treatments compared to the control. The result obtained from this study is thus likely due to the quantity added. According to Windisch et al.[46] supplementation levels of products to poultry diets vary between 0.1 and 40 g/kg for dried products and plant extracts. In addition, phytobiotics (PSP) are substance added to feed to improve the safety, flavor, taste, and other qualities of the feed[47 48] and do not add significantly to the main nutrients of the feed.

Weight gain in poultry remained ($P > 0.05$) with increasing level of PSP in the diet from 0.5% to 1.0 % (Table 2). Usually, increase weight gain by broiler chicks fed additive containing diet observed in this study could be attributed to the fact that herbal plants provide compounds that enhance digestion and absorption of nutrients in diets, leading to improved growth of chicks[49]. The result of this study was not in concordance with that of Mohamed et al.[50], and Arshad et al.[51] who reported significant ($P < 0.05$) increase in live body weight of broiler chicks fed ginger-supplemented diet.

Generally, better feed conversion ratio values were obtained in chicks fed all treatments, but getting poor with increase of dietary PSP up to 1 % PP. Chicks fed diet containing 0.5 % PP had better feed conversion ratio. Better feed conversion ratio of the broiler chicks fed diet containing PSP additive could be attributed to the antibacterial properties of these additive, which may have resulted in better absorption of the nutrients present in the gut and finely leading to improvement in feed conversion ratio of the treatments group[52].

The broiler chicks fed PSP diets showed better feed intake compared to the control (Table 2). Feed intake was significantly ($P < 0.05$) the highest in 1% PSP. Feed intake of broilers in this study showed that feeding PSP resulted in dose-related effect on palatability[53 54]. Better feed intake of broiler chicks fed PSP diets could be attributed to an improvement in the oganoleptic characteristics of feed as a result of adding PSP. Arshad[51], and Herawati[55], reported similar results in feed intake when broiler chicks were fed diet containing ginger powder as phytobiotics additives.

Blood acts as a pathological reflector of the status of animals exposed to xenobiotics and other extreme conditions[56]. As reported by Isaac et al.[57] animals with healthy blood composition are likely to show good performance. Laboratory tests on the blood are vital tools that help detect any deviation from normal health status in animals[58]. The examination of blood gives the opportunity to investigate the presence of several metabolites and other constituents in the body of animals and also play vital roles in the physiological, nutritional and pathological status of an organism[59 60]. According to Afolabi et al.[61], changes in haematological parameters are often used to determine status of the body and stresses due to environmental, nutritional and/or pathological factors.

Haematology parameters such as MCV, HCT% and PLT did not significantly ($P > 0.05$) differ across all the treatments (Table 4.5). Generally, inadequate intake of energy and protein adversely affect RBC, HGB, MCV and HCT or packed cell volume (PCV) concentration which indicates anaemia[62 63]. RBC and HGB values recorded were higher ($P > 0.05$) in the PSP- free diet group compared to 0.5 % PSP supplemented group and it became lower with the increase in PSP up to 1 % PSP. The increase observed in broiler chicken fed 0 % PSP diet might be due to the greater oxygen demand for activity[64]. Moreover, Palomeque et al.[65] reported that increased haemoglobin content per unit of volume of blood may be a reflection of decreased volume of blood per unit body weight.

The normal values of HCT obtained in the present study indicated that the PSP added to chicken diet increased the availability of protein, energy and the degradation of anti-nutritional factors. This according to Cary, et al.[66] could consequently improve broiler performance. This confirmed that haematological traits, especially HCT and HGB were correlated with the nutritional status of the animal[67] and agreed with Oyawoye, and Ogunkunle[68], who stated that PCV is an index of toxicity in the blood and high levels usually suggest the presence of toxic factors which has adverse effect on blood formation.

Blood parameters such as WBC, LYM, MCH, MCHC and MPV did not significantly ($P > 0.05$) differ across all the treatments (Table 4.6). The normal values of WBC, platelets, and MCHC recorded for broiler chicks fed PSP diet may be indication that the bone marrows of the broiler chickens were properly functional and also revealed the absence of macrocytic and hypochronic anaemia[69].

Normal value of WBC recorded for broiler chickens fed PSP diet is probable due to the ability of PSP to cause destruction or impaired production of white blood cells, probably by affecting the production of regulatory factors involved in haematopoesis[70]. However, PSP has been shown to have antioxidant properties which could equally suppress production of white blood cells[71].

The result of MCH, which is the average amount of HGB in each RBC measured, followed the same pattern as MCV. The MCV and MCH values obtained in broiler chickens fed PSP diet and PSP free diet in this study fall within the normal range of 90 to 140 fl and 16 to 65pg[72 73]. The result of this study is in line with that reported by Tuleun et al.[74] in broiler chickens and Adenkola et al.[75] in rabbits that nutrient is an important factor in haemopoesis. .

Hepatic and renal function biomarkers of broiler chickens fed graded level of PSP for 6 weeks starter period showed AST, ALT and ALP values not significantly varied ($P > 0.05$) across all the treatments (Table 4.7). Notably, the values of AST and ALT obtained in broiler chicks fed PSP containing diet were lower when compared to the control diet (0 % PSP) which, however, still falls within the normal range in chickens; from 1-37 µL[25]. This likely influenced the ability of birds to withstand the effect of anti-nutritional factors (if any) which could cause liver damage[76]. This was similar with the findings recorded by Ekpenyong and Biobaku[77], who stated that the values of AST and ALT are normally low in blood but becomes high when there is occurrence of liver damage by toxic substances.

Similar values of AST and ALT that were not significantly varied (p>0.05) in this study for the control (0% PP) relative to treatments are indicative of normal liver and kidney functions[78 79] that did not worsen by addition of more PSP.

The result of the serum metabolic parameters; serum albumin, creatinine, urea and total protein of treatments (0.5% PP and 1% PP) (Table 4.8) were similar with that for the control (0% PP). High urea and creatinine values are a measure of especially muscle amino acid degradation[80], and early pointer to depressed liver and kidney functions[81].

Creatinine values of broiler chickens fed PSP containing diet were numerically lower but not significant (P > 0.05) compared to the control. Elevated creatinine value in broiler chickens fed PSP-free diet may suggest depletion of tissue creatinine phosphate and this may adversely affect the muscle mass[82 83]. This could imply that there was slightly better protein metabolism and utilization in the treatment group than the control. Therefore, PSP can be used with confidence in broiler diet to provide adequate nutrition.

The result of the activities of Superoxide Dismutase and Catalase as antioxidant enzymes were significantly (P<0.05) elevated in treatment with PSP when compared to the control (Table 4.9). The values of SOD and CAT increased significantly (P<0.05) with increasing levels of PSP. Oxidative stress is the potential mechanism that overwhelms the primary antioxidant defense of the body. Antioxidants function by suppressing the formation of reactive oxygen species (ROS) and reactive nitrogen species (RNS), affecting enzyme activities[29]. SOD is one of the crucial antioxidant defenses of the body, and only enzyme family with activity against superoxide radicals. SOD enzymes are present in almost all aerobic cells and in extracellular fluids[84].

Catalases are enzymes that catalyses the conversion of hydrogen peroxide to water (H_2O) and oxygen (O_2), using either an iron or manganese cofactor[85 86]. This protein is localized to peroxisomes in most eukaryotic cells[87]. Therefore, increased values recorded as the dietary level of additive was increased from 0.5% PP to 1% PP could be attributed to antioxidant effect of papaya seeds which stimulated the activities of both superoxide dismutase and catalase enzymes.

Conversely, lipid peroxidation profile decreased significantly (p<0.05) in treatments with PSP when compared to the control (Table 4.9). Lipid peroxidation is the oxidative degradation of lipids. It is the process in which free radicals steals electrons from the lipid in cell membranes, resulting in cell damage[88]. In this study, lipid perodixidation values tended to decrease as the dietary level of PSP was increased from 0.5% PP to 1% PP. This may be attributed to the antioxidant effects of PSP which are capable of neutralizing reactive oxygen species before lipid peroxidation is initiated[89]. Similar result was reported by Antache, et al.[90] who showed that the supplementation of Nile tilapia feed with rosemary, sea buckthorn and ginger in 1% concentration led to reduction in malondialdehyde concentration, and increased total antioxidant capacity from various tissues.

V. Conclusion

From the results of this experimentation, feeding broiler chickens with diets mixed with PSP significantly and positively reduced serum lipid peroxidation profile without negatively affecting feed efficiency, growth performance and serum biochemical parameters. Therefore, PSP could be supplemented in feed as phytobitic additives because it has potentials as antioxidant when supplemented in the diets of broiler chickens.

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