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HAEMATOLOGICAL AND SERUM BIOCHEMICAL RESPONSES OF GROWING PIGS TO *SPONDIAS MOMBIN* LEAF MEAL SUPPLEMENTATION

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ABSTRACT

Pig production plays an essential function in food security, remedy of poverty, and employment in Nigeria. However, feed given to growing pigs determines its growth and overall health status. Therefore, this study was aimed at evaluating the overall physiological performance of the grower pig-fed *Spondias mombin* leaf meal (SMLM). A total of 24 weaned pigs weighing between 9.37kg – 12.18kg were allotted to 4 treatments each having 2 replicates per treatment in a completely randomized design (CRD). Treatment 1 with no SMLM served as the control diet, and treatments with 5, 6 and 7% of SMLM for treatments 2, 3 and 4 respectively. There was a significant difference ($p<0.05$) in Packed Cell Volume (PCV) and Red Blood Cells (RBC) among treatments with values in male pigs in T4 showing the highest mean value. Serum indices of the female pigs were all similar to those of MCHC with pigs in T3 and T4 being the highest and similar values. For the leukocytic indices profile of the pigs, there was a significant difference ($p<0.05$) among mean values of WBC and platelets in both male and female animals with T4 showing the highest mean values for both parameters, which can be attributed to variation in SMLM level in respective diets. All other leukocytic indices values assessed (heterophils, monophils, lymphocytes) were similar. The presence of basophils was not observed during serum examination of blood samples. Based on the outcome of this study, *Spondias mombin* leaf meal had no deleterious effect on the haematological and serum biochemical indices of growing pigs up to 7% dietary level. It was therefore, concluded that 7% dietary level of inclusion is safe for growing pigs.

Keywords: Physiological status, *Spondias mombin* leaf meal, Supplementation, blood profile.

INTRODUCTION

The physiology of farm animals is influenced by multiple factors, with nutrition being a critical one (Ajao *et al*., 2013). Nutrition is the most crucial factor affecting livestock production in Nigeria. The main militating factor in the production of pig products is the high cost of feed ingredients as reported (Okorie *et al.*, 2016) that quality feeds are expensive because most of the constituent feed materials such as maize, soya bean, fish meal and groundnut cake are utilised not only by the animals but by humans and industries as well. Blood analysis is a rapid and readily available tool for evaluating the nutritional status of animals undergoing feeding trials. This is because dietary components induce measurable changes in blood composition, providing a reliable indicator of long-term nutritional status (Etim *et al*., 2014a; Lim *et al*., 2021). Furthermore, the haematological and serum biochemistry profiles of animals are significantly shaped by various factors, particularly the nutritional quality of their feed. Understanding how different feed ingredients

impact the haematological indices of swine is crucial in determining their suitability for use in pig feed formulations.

Haematological examination has been performed for various reasons; as a screening procedure to examine the general health of the animal, to assess the nutritional status of the animal, as an adjunct to infection and to evaluate the progress of certain disease conditions (Friday *et al*., 2014) and to distinguish between normal and stress conditions of the animal (Olabanji *et al*., 2007). Haematology values provide baseline information for comparison in conditions of nutrient deficiency, physiology and health status of farm animals (Nse Abasi *et al*., 2014) and help in providing information on the relationship between blood characteristics and environment (Ovuru and Ekweozor, 2004, Isaac *et al*., 2013).

The sustainability of modern animal production relies heavily on the strategic and innovative use of feed additives to produce affordable and nutritious food on a large scale. Feed additives, particularly medicinal plants, play a crucial role in maintaining animal health, enhancing feed quality, and promoting growth performance. As noted by Duduku *et al.*, 2011; Alagbe *et al*., 2018, medicinal plants possess various bioactive properties, including anti-inflammatory, antiseptic, antibacterial, anthelmintic, and antioxidant activities, which can effectively combat microorganisms, treat gastrointestinal issues, and promote overall well-being Adesuyi *et al*., 2012; Abdi-Hachesoo *et al*., 2011; Arkan *et al*., 2012.

Spondias mombin Linn, a fructiferous tree that belongs to the family Anacardiacae is common in Nigeria and several other tropical forests of the world with high genetic variability among populations (Ayoka, *et al*., 2008). According to Burt (2004), herbs and spices have been found to exhibit potent antimicrobial properties in vitro against pathogens, making them a viable alternative to antibiotic growth promoters in animal feed. These plant-based feed additives have been shown to offer a range of benefits, including enhanced animal performance, improved nutrient availability, and reduced toxicity compared to organic chemicals. Additionally, they do not leave unwanted residues and can serve as dietary supplements.

Herbs, typically derived from leaves, contain phytochemicals, which are chemical compounds produced by plants as part of their metabolic processes. As noted by Dalle Zotte *et al*. (2016), phytochemicals can be classified based on their therapeutic values, including antibacterial, antifungal, anti-inflammatory, antiulcer, antioxidant, antiviral, anticancer, and immune-stimulating properties. Furthermore, phytochemicals can be prepared in various forms, such as tinctures, decoctions, macerations, syrups, inhalations, and infusions.

MATERIALS AND METHODS

Experimental location

This experiment was carried out at the piggery unit of Teaching and Research Farm, University of Ibadan, Nigeria situated at the latitude $07^{0}26^{0}$ N and longitude $03^{0}54^{0}$E with elevation ranging from 150mm-275mm above sea level.

Procurement and processing of the experimental materials

The leaves of *Spondias mombin* were harvested and air dried to constant weight under room temperature for 7 days and milled in a hammer mill. The sample of the leaf meal was subjected to proximate analysis according to (AOAC, 2010).

Experimental animals and management

Twenty-four (24) 3-months old pigs with an average weight of 9kg were used. Ivermectin was administered to the animals and lubricating oil was applied against external parasites. They

were allowed 14 days to adjust to feed and confinement before the actual start of the experiment.

Experimental diet

Four dietary treatments containing levels of *Spondias mombin* dry leaf meal at 0, 5, 6 and 7% inclusion rates were formulated and designated as T1, T2, T3, and T4. Respectively. Treatment 1 as the control had no *Spondias mombin* leaf meal.

Experimental design

The pigs were randomly allocated to four dietary treatment with six pigs per treatment in a completely randomized design. The feeding trials lasted for three months. The pigs were offered feed at 5% of their body weight. All pigs were weighed at the beginning of the experiment and every week thereafter in order to maintain the pre-determined level of 5% feeding of the animals' body weight in the period of study.

Blood sample collection

About 5ml of blood (2mL for haematology and 3mL for serum analysis) was taken from the cranial vena cava of the pigs using needle and syringe.

Haematology

The blood samples were placed in specimen bottles with EDTA (Ethylene Diamine Tetra Acetate) for haematological studies. Blood was analysed for Packed cell volume, Haemoglobin, Red blood cell, White blood cell, Mean cell volume, Mean haemoglobin as outlined (Ochei and Kolhatker 2000).

Serum biochemical analyses

Blood samples placed in the specimen bottles without EDTA were used to analyse the selected serum biochemical indices such as glucose, total protein, albumin, and creatinine as outlined by Ochei and Kolhatker (2000).

Statistical Analyses

Data were analyzed using one-way analysis of variance (Statistical Analysis System, version 9.2, 2005) and the means were separated using the Duncan's Multiple Range Test. Values were considered statistically significant at $p<0.05$

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RESULTS

Table 1: Proximate Analysis of *Spondias mombin* Leaf Meal

Parameters	Fresh leaves		Milled leaves	
% Crude protein	8.75	8.69	14.07	14.20
%Ash	13.11	13.20	12.48	12.35
%Crude fibre	10.85	11.10	9.55	9.70
%Ether extract	2.90	3.10	3.60	3.80
% Dry matter	92.25	92.10	93.31	93.02
ME(Kcal/kg)	2846.815	2843.92	2952.17	2964.47

ME Metabolisable Energy

Table 2: Percentage Composition of Experimental Diet (Dry Matter Basis)

	Treatments			
Ingredients	T1	T2	T3	T4
Maize	27.00	22.00	21.00	20.00
SMLM	0.00	5.00	6.00	7.00
Palm oil	3.00	3.00	3.00	3.00
SBM	17.00	17.00	17.00	17.00
Wheat offal	22.00	22.00	22.00	22.00
PKC	25.00	25.00	25.00	25.00
Fish meal	1.00	1.00	1.00	1.00
DCP	2.50	2.50	2.50	2.50
Oyster shell	1.50	1.50	1.50	1.50
Premix	0.25	0.25	0.25	0.25
Salt	0.25	0.25	0.25	0.25
Lysine	0.30	0.30	0.30	0.30
Methionine	0.20	0.20	0.20	0.20
Total	100.00	100.00	100.00	100.00
Calculated analysis				
ME(Kcal/kg)	2527.40	2512.73	2509.72	2506.71
Crude protein	17.00	15.02	18.28	18.54
Crude fibre	7.50	8.22	18.36	8.50

SMLM = Spondias mombin leaf meal
SBM = Soy bean meal
PKC = Palm kernel cake
DCP = Dicalcium Phosphate
ME = Metabolizable energy

Table 3: Haematological indices of male pigs fed varying level of *Spodias Mombin* Leaf Meal

	Treatments				
Parameters	**T1(0%)**	**T2(5%)**	**T3(6%)**	**T4(7%)**	**SEM**
PCV(%)	34.50^{ab}	38.50^{ab}	33.50^{b}	41.00^{a}	1.11
Hb (g/dL)	11.00	12.30	10.80	12.55	1.12
RBCx10^{6}µL	5.59^{ab}	6.07^{ab}	5.48^{b}	6.75^{a}	0.18
MCV	61.64^{b}	63.38^{a}	61.19^{b}	60.68^{c}	0.41
MCHC	31.88	31.95	32.18	30.48	0.39
MCH	19.65	20.25	19.68	18.49	0.27
WBCx10^{3}	6.32^{b}	5.10^{c}	5.05^{c}	7.67^{a}	0.04
Plateletsx10^{3}	93.00^{ab}	76.00^{b}	103.00^{a}	77.50^{b}	3.17
Lymphocyte(%)	68.00	69.00	63.50	69.00	0.88
Heterophils(%)	28.50^{b}	28.00^{b}	33.00^{a}	27.50^{c}	1.02
Monophils(%)	1.50	2.00	1.50	2.00	0.21
Eosinophils(%)	2.00	1.00	2.00	1.50	0.25

^{abc} means within the same row with different superscripts are significantly different ($p < 0.05$).

PCV = Packed cell volume
Hb = Haemoglobin
RBC = Red blood cells
MCV = Mean corpuscular volume
MCHC = Mean corpuscular haemoglobin
MCH = Mean corpuscular haemoglobin
WBC = White blood cells
SEM = Standard error of mean

Haematological indices of male pigs fed varying level of *Spodias Mombin* Leaf Meal

Table 3 shows the haematological indices of the male pigs i.e packed cell volume, haemoglobin concentration, red blood cells, mean corpuscular volume, mean corpuscular haemoglobin concentration, mean corpuscular haemoglobin. MCHC shows no significance difference, this trend is also observed for other parameters except PCV, RBC, MCV, WBC, Heterophils and Platelets. For PCV, T4 and T2 are significantly higher compare to T3 and T1. T3 and T1 are significantly higher compare to T4 and T2 for platelets.

Table 4: Haematological indices of female pigs fed varying level of *Spondias Mombin* Leaf Meal

	Treatments				
Parameters	**T1(0%)**	**T2(5%)**	**T3(6%)**	**T4(7%)**	**SEM**
PCV (%)	34.50^{c}	39.50^{b}	38.50^{b}	40.50^{a}	1.52
Hb (g/dL)	11.10^{c}	12.80^{b}	12.80^{b}	13.30^{a}	1.64
RBCx10^{6}µL	5.66	6.55	6.50	6.82	0.93
MCV	61.19	60.29	59.23	59.32	0.31
MCHC	32.03^{b}	32.41^{ab}	33.24^{a}	32.85^{ab}	0.17
MCH	19.59	19.53	19.69	19.49	0.03
WBCx10^{3}	5.95^{b}	4.57^{b}	5.45^{b}	7.70^{a}	0.37
Plateletsx10^{3}	64.00^{c}	104.00^{a}	100.00^{a}	93.50^{a}	18.66
Lymphocyte(%)	66.00	71.00	68.50	71.50	1.36
Heterophils(%)	33.00	25.50	28.0	25.50	1.55
Monophils(%)	1.00	1.50	1.50	1.50	0.00
Eosinophils(%)	0.00	2.00	2.00	2.00	0.33

^{abc}Means within the same row with different superscripts are significantly different ($p< 0.05$).

PCV = Packed cell volume
Hb = Haemoglobin
RBC = Red blood cells
MCV = Mean corpuscular volume
MCHC = Mean corpuscular haemoglobin
MCH = Mean corpuscular haemoglobin
WBC = White blood cells
SEM = Standard error of mean

Haematological indices of female pigs fed varying level of SMLM

Table 4 shows the haematological indices of the female pigs. PCV, Hb, WBC and Platelets all were significantly different ($p< 0.05$) while there was no significance difference for other parameters. Treatments 2 and 3 are significantly higher ($p< 0.05$) than T4 and T1 for platelets.

Table 5: Serum biochemical indices of male pigs fed varying level of *Spondias Mombin* Leaf Meal

	Treatments				
Parameters	**T1(0%)**	**T2(5%)**	**T3(6%)**	**T4(7%)**	**SEM**
Glucose(mmol/L)	58.29[a]	49.44[c]	49.10[c]	55.34[b]	1.77
Total protein(g/dL)	6.31[ab]	5.50[b]	6.45[ab]	7.08[a]	0.19
Creatinine(mmol/L)	28.33[b]	26.67[c]	32.33[a]	29.67[b]	1.72
Albumin(g/L)	3.35	2.98	3.18	3.27	3.82
Globulin(g/l)	2.97	2.52	3.27	3.82	0.21
Alb:Glo	1.13	1.34	0.97	0.89	0.10

[abc]Means within the same row with different superscripts are significantly different ($p< 0.05$).

Alb:Glo = Albumin:Globulin

SEM = Standard error of mean

Table 5: Serum biochemical indices of male pigs fed varying level of SMLM

Presented in Table 5 are the serum biochemical indices of male pigs. Glucose, Total protein and Creatinine all show significance difference while all other parameters showed no significance difference. T4 and T1 showed a higher significant difference compared to T3 and T2 for creatinine.

Table 6: Serum biochemical indices of female pigs fed varying level of *Spondias Mombin* Leaf Meal

	Treatments				
Parameters	**T1(0%)**	**T2(5%)**	**T3(6%)**	**T4(7%)**	**SEM**
Glucose(mmol/L)	46.32[b]	49.17[a]	42.30[c]	44.09[b]	2.07
Total protein(g/dL)	6.02[b]	6.32[ab]	7.15[a]	5.81[b]	0.17
Creatinine(mmol/L)	24.67[b]	29.00[a]	28.33[ab]	29.67[a]	1.25
Albumin(g/L)	3.25	3.31	3.42	3.23	0.04
Globulin(g/l)	2.77[ab]	3.00[ab]	3.73[a]	2.58[b]	0.16
Alb:Glo	1.19	1.16	0.94	1.26	0.05

[abc]Means within the same row with different superscripts are significantly different ($p< 0.05$).

Alb:Glo = Albumin:Globulin

SEM = Standard error of mean

Table 6: Serum biochemical indices of female pigs fed varying level of SMLM

Table 6 shows the serum biochemical indices of female pigs, there was significance difference for Glucose, Total protein, Creatinine and Globulin. There was no significance difference for Albumin and Alb;Glo.

DISCUSSION

Haematology

Erythrocytic Indicies

Serum biochemical constituents reflect the physiological responsiveness of the animals to its internal and external environment, which include feed and feeding (Esonu *et al*., 2001). From this study, all PCV values measured are within standard range as referenced by Thorn (2000). Packed Cell Volume concentration is important in assessing blood profile as it is used in determination of, conjunction with haemoglobin concentration, mean corpuscular haemoglobin concentration of examined animals to assess level of dehydration and anaemia as well iron deficiencies. Causes of variation in values may be indicative of varying sources. However, it is obvious from the current study that SMLM inclusion in diet support blood cells proliferation as animals in T4 showed highest values of PVC. Chineke *et al.*, (2006) reported that apart from genotype, age, sex, differences in haematological indices may be caused by nutritional, environmental and hormonal factors. Management is another factor that affects PCV of animals. Olayemi *et al*., (2000) conducted a study to determine the haematology of the West African Dwarf (WAD) sheep under intensive and extensive management systems in Nigeria. The intensively reared animals showed higher PCV, Hb concentration and MCV than those under extensive management. This might be connected with animals picking toxicants and anti-nutritional factors in course of scavenging. This supports the opinion of Olafedehan *et al*., (2010) blood act as a pathological reflector of the status of exposed animals to toxicant and other conditions.

Leukocytic Indices

Our entire environment is habited by various microorganisms. Some are harmful, while others are not. The presence of microbes poses constant challenge to the animal body, giving rise for continuing defence mechanism to fight of effects of pathogens. The white blood cells, stands as chief cells of defence in the mammalian body. This experiment reveals a significant difference in total WBC value measured as well as platelets which serve in repairs of damaged cells. Although, mean differences in lymphocyte, heterophils, monophils and eosinophils are not significant, total WBC shows significant increase in values with animals in T4 showing highest values. Phytogenic feed additive, such as *Spondias mombin* leaf meal, are plant derived compounds added to animal's diet to improve its productivity via amelioration of feed properties, improvement of nutrient digestibility, absorption and eliminations of pathogens in the gut (Ayoka *et al*., 2005).

Serum Biochemistry

Serum Metabolites

Mean values for glucose, total protein, creatinine, albumin and globulin of male and female pigs assessed in this study all fall within the ranges referenced in RAR (2009). Significant difference in total protein concentration in both sexes increased progressively with increase in dietary levels, indicating potential of SMLM as an additional protein source in feedstuffs in animal diets. Plasma proteins serve as a source of nutrition for tissues. A dynamic equilibrium exists between the proteins of plasma and those of tissue (Sokunbi, 2007). High total protein concentration aids the synthesis of albumin and globulin within the body. Sanchez-Monge *et al*., (2004) reported increased globulins to be implicated in liver damage, chronic infections and kidney dysfunction. Akinmutimi (2004) reported that low levels of globulin affect the animal's ability to fight disease. Globulin

formation is essential for the synthesis of functional immune proteins. Inclusion of SMLM in the diet caused a significant difference in globulin concentration in female pigs with 5% inclusion resulting in the highest mean value.

Albumin to globulin ratio

The health status of animals as well as the nutritive values of feedstuffs can also be assessed from utilization for physiological process availability. As a composite biomarker for monitoring inflammation and nutritional status, distortions in the A/G ratio show challenging conditions which may result from management or disease (Chen *et al.* 2022). It is evident from this study that the A/G ratio was slightly above the 1:1 approximate value reported by Wong *et al*. (2013). Although not significant, the mean values of pigs in T3 both male and female, were less than 1. This may be as a result of adjusting to increasing levels of SMLM in feed by male animals while the females metabolise it better for biochemical processes.

CONCLUSION

This experiment shows that graded inclusion of *Spondias mombin* leaves meal did not have any adverse effect on the physiological status of the animals. Further studies will be necessary with higher inclusion levels of SMLM to establish the optimal level of inclusion of SMLM for weaned pigs and other production stages of pigs. Further research is also necessary to elucidate the underlying mechanisms of its effects on haematology and serum biochemistry, as well as its potential interactions with other dietary constituents.

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