

# Haematological Indices and Growth Performance of Broilers Fed *Annona Muricata* Leaf Meal

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**Abstract-** *This experiment was conducted to investigate the haematological indices and growth performance of broilers fed diets containing *Annona muricata* leaf meal for eight weeks. A total of 60 unsexed day-old broiler birds were obtained from Agrited farms and were allotted to four treatments (T1, T2, T3 and T4) and replicated three times with five birds per replicate in a Completely Randomized Design. The birds in T1, T2, T3 and T4 were fed with diets containing 0, 2, 4 and 6% *Annona muricata* leaf meal. The haematological parameters determined were packed cell volume, red blood cell count, haemoglobin, white blood cell count, mean cell volume, mean cell haemoglobin estimation, mean cell haemoglobin concentration, and platelet count. Growth parameters measured were the initial weight, final weight gain, average daily feed intake, average daily weight gain, total feed intake and feed conversion ratio. The data collected was analyzed using one-way Analysis of Variance (ANOVA). The result of the experiment showed that there were no significant differences ( $P>0.05$ ) in all the haematological indices and they were within the normal range with the exception of the WBC. There were significant differences ( $P<0.05$ ) in the growth performance with the exception of the initial weight. Thus, *Annona muricata* leaf meal is not recommended to be included in diets of broilers above 2% levels of inclusion as its inclusion does not have any economic benefit and decreased the growth performance due to its high fiber content and phytochemical properties.*

**Index Terms-** *Annona muricata, broiler, growth, haematology, leaf meal.*

## I. INTRODUCTION

Broilers are sensitive to environmental effects especially feed during their growth and development. Many plant parts such as the leaves have shown their capacity to influence the growth performance, haematological and serum biochemistry of broilers at different stages of their growth (Jiwuba et al., 2017).

The effects of these plant parts may be different due to the synergistic or antagonistic effects of the plant parts when tested in combination with other plant parts. The effects of these plant parts depend on their proximate composition, mineral and phytochemical composition. Most of the plant parts promote growth by suppressing the activities of pathogens in the body of these animals. Others promote growth by providing essential nutrients necessary for growth.

Feed remains the most expensive factor of production in every broiler production enterprise, contributing up to 70% of the cost of production (FAO, 2016). Feed ingredients are expensive and have continued to increase in price due to the increasing human population. There is also greater demand for animal product necessitating an increase in the production capacity in order to meet the minimum requirements for animal protein as stipulated by Boluwaji et al., (2024). Bridging the protein demand gap will need the efforts of the government, broiler farmers and researchers alike. Government has introduced many interventions geared towards reducing the cost of feed ingredients like the Anco borrowers programme in Nigeria etc. Farmers are also making some efforts in this direction. Researchers on the other hand have made a lot of commitments through research and advocacy programmes with the effort of reducing the cost of feed ingredients and at the same time reduce the cost of broiler production. In line with this, *Annona muricata* leaf meal efficacy in promoting the growth and haematological indices of broiler will be investigated.

*Annona muricata* leaf meal (AMLM), Sour soup leaves is one of natural ingredients that contains many active substances that can prevent disease and

improve productivity of livestock. AMLM contains flavonoid, tannin, phytosterol, oxalate calcium, alkaloid, acetogenins and steroid (Suranto, 2011). Flavonoid is antioxidant that protects cell structure, improves efficacy of vitamin C and serves as anti-inflammation, antibacterial and antibiotic. Flavonoids such as quercetin and epicatechin have antidiarrheal effect and can improve natural body immune response (Russell and Taylor 1998). Saponin for non-ruminant feed like broiler is known to improve growth, feed efficiency and meat quality. Tannin derived from several plant extracts can prevent colonization of parasites, bacteria, protozoa and viruses in digestive tracts; therefore, it is a common traditional medicine for diarrhea and dysentery (Maertens and Štruklec, 2006).

Leaf meal such as *Annona muricata* has the potential for improving the growth and hematological properties of broilers due to its phytochemical properties (Artawiguna et al., 2023). However, the efficiency of its utilization in poultry is not properly established. Hence the need to evaluate the haematological and growth performance of broilers fed diets containing *Annona muricata* leaf meal.

## II. MATERIALS AND METHODS

### Experimental Site

This study was conducted at the experimental poultry farm of Nnamdi Azikiwe University, Awka, Anambra state in Nigeria. Awka is located at the tropical rain forest zone at Latitude 6° 12' 15.00" and longitude 7° 04' 2.40". The mean average annual temperature is 27.0°C while average annual rainfall is 1828mm with a bimodal rainfall pattern.

### Collection and Preparation of Experimental Materials

Fresh leaves of AMLM were sourced within Awka, Anambra state. The leaves were harvested from the tree, separated from the stalk, air dried for about 3 weeks and milled in a hammer mill to produce sour soup leaf meal (SSLM). Processed full fat soya bean was purchased from New market, Enugu State.

### Experimental Diets

The AMLM and full fat soya bean were used together with other ingredients to formulate four diets for both the starter mash and the finisher mash. The diets contained varying levels of Sour soup leaf meal (0%, 2%, 4% and 6%) and other ingredients were included to make the diet isonitrogenous and isocaloric. The diets were designated T1 (0%SSLM), T2 (2%SSLM), T3 (4% SSLM) and T4 (6% SSLM) respectively. Table 1 presents the percentage feed stuff at various levels of inclusion of AMLM in the formulated broiler starter diet.

Table 1: Percentage feedstuff composition in the broiler starter diet

Ingredients Inclusions	Varying Levels of				
	%	T1 (0%)	T2 (2%)	T3 (4%)	T4 (6%)
Crude Protein	8.9	40	40	40	40
Maize Spent Grain	20	24.5	23.5	21.5	19.5
Pkc	16	3	3	3	3
Wheat Offal	15	3.5	2.5	2.5	2.5
Fullfat Soybean	46	22	22	22	22
Lysine	95.4	0.5	0.5	0.5	0.5
Methionine	58.4	0.5	0.5	0.5	0.5
Toxin Binder	-	0.4	0.4	0.4	0.4
Salt	-	0.4	0.4	0.4	0.4
Oyster Shell	-	2	2	2	2
Bone Meal	56.8	3	3	3	3
AMLM	17.1	0	2	4	6
Vitamin Premix.	-	0.2	0.2	0.2	0.2
		%cp=2 2.05	%cp=2 2.05	%cp=2 2.0	%cp=2 2.2

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Table 2 presents the percentage feed stuff composition at various levels of inclusion of AMLM at finisher stage.

Table 2: Percentage feed stuff in the broiler finisher diet

Ingredients Inclusions of AMLM	Varying Levels of				
	%Cr ude Prot ein	T1 (0%)	T2 (2%)	T3 (4%)	T4 (6%)
Maize	8.9	40	40	40	40
Spent Grain	20	8	6	5.5	5
Pkc	16	12	13	12.5	11
Wheat	15	18	17	16	16
Offal					
Full Fat	46	14	14	14	14
Soybe an					
Lysine	95.4	0.5	0.5	0.5	0.5
Methi onine	58.4	0.5	0.5	0.5	0.5
Toxin	-	0.4	0.4	0.4	0.4
Binder					
Salt	-	0.4	0.4	0.4	0.4
Fish	66	3	3	3	3
Meal					
Bone Meal	56.8	3	3	3	3
AML	17.1	0	2	4	6
M	5				
Vitami n	-	0.2	0.2	0.2	0.2
Premi x.					
		%cp= 20.49	%cp= 20.63	%cp= 20.64	%cp= 20.64

Experimental Design

Sixty broiler chicks were randomly assigned to the four dietary treatments. Each treatment was replicated thrice with 5 birds per replicate in a completely randomized design (CRD). The data collected was subjected to Analysis of variance (ANOVA) using SPSS analytical package and differences between means were separated using Duncan multiple range test (Duncan, 1955) at 5% probability level.

### III. MANAGEMENT OF THE EXPERIMENTAL BIRDS

#### Brooding and housing of the birds

The brooding pen was washed with detergent and water and disinfected using Izal. It was left for seven days before wood shavings were placed on the cemented floor of the pen in preparation of the bird's arrival. A total of sixty (60) unsexed broiler chicks from Agrited were used for the experiment. The broiler chicks were brooded for four weeks after which they were reared for another four weeks. Each replicate pen was equipped with feeding trough, drinkers and kerosene lanterns as source of brooding heat and light. They were weighed individually and randomly assigned to various dietary treatments. Each treatment level had three replicates. The feed and clean drinking water were provided at all times. Management practices for optimal growth and development such as timely vaccination, feeding, Sanitation as well as pest and disease control were carried out as at when due for accuracy of results.

#### Feeding of the Birds

From day old to the 28th day, the birds were fed with experimental starter feed after which they were introduced to the experimental finisher feed from 29th day to the 56th day. The feeds were formulated by a nutritionist to ensure they were isocaloric and isonitrogenous so that the only source of variation will be the various levels of inclusion of sour soup leaf meal. The birds were fed *ad libitum* with the experimental diet which was provided in a conical feeder and long feeder during the course of the experiment. Fresh, clean drinking water was provided using the medium size drinkers.

### Vaccination / Medication

Diseases of poultry were prevented through the use of vaccine. The vaccines given and the time of administration were given as in the vaccination schedule in table 3.

Table 3: Vaccination schedule for broilers

Age in days	Type of vaccine	Disease	Route of administration
8	Gumboro disease vaccine (1st dose).	Gumboro.	Oral through drinking water.
14	New castle disease vaccine-Lasota (1st dose).	New castle disease.	Oral through drinking water.
21	Gumboro disease Vaccine (2nd dose).	Gumboro.	Oral through drinking water.
28	New castle disease vaccine-Lasota (2nd dose).	New castle disease.	Oral through drinking water.

### Sanitation and Hygiene

The rice husk was changed regularly usually once every week from week 2 to week 8. This was done to prevent the accumulation of manure which can be a source of disease infection to the birds. The drinkers and feeders were washed thoroughly everyday with detergent and clean water. Bathroom slipper was provided at the entrance of each pen to prevent infection from one pen to the other. For the purpose of convenience, the chicks were fed with shallow feeders and drinkers in their first four weeks. The feeders were later suspended using a rope after their fourth week and drinkers were raised to avoid fouling with droppings and litter materials. Also, at the entrance of the farm house, water containing disinfectant was provided for anybody entering the poultry house to dip their feet. This was done to reduce the risk of disease infection and outbreak in the poultry house.

### Data Collection

Daily feed intake was calculated using feedback mechanism while weight gain was measured every week using weighing balance. Blood samples were taken from one bird per replicate making a total of 3 birds per treatment at eight weeks by inserting a sterile needle into the wing vein of the birds and extracting 2mls of blood each. The blood samples were placed in previously labeled and sterilized sample tubes containing EDTA for hematology analysis. The blood samples were immediately transported to the laboratory for analysis.

### Haematological Technique

Differential WBC counts were made on monolayer blood films, fixed and stained with Giemsa-Wright's stain. Total red blood cell (TRBC) and total white blood cell count (TWBC) were determined by a manual method using hemacy-tometer (Campbell, 1995). Packed cell volume (PCV) was measured by a standard manual technique using microhematocrit capillary tubes and centrifuged at 2500 rpm for 5 min. Hemoglobin concentration (Hb) was measured by Cyanmeth-emoglobin method. Erythrocyte indices (mean corpuscular volume (MCV), mean corpuscular hemoglobin (MCH) and mean corpuscular hemoglobin concentrations (MCHC) were calculated from TRBC, PCV and Hb (Ritchie et al., 1994), respectively.

### Data Analysis

All the data collected were subjected to analysis of variance (ANOVA) using the Statistical Package for Social Sciences (SPSS). Significant difference among treatment means were separated using Duncan multiple range test at 5% significance level.

## IV. RESULTS

Haematological indices of broilers fed diets containing *Annona muricata* leaf meal.

The haematological indices of broiler fed diets containing AMLM is presented in table 4. The table shows that all the treatment means of the various haematological indices were not significantly different ( $P > 0.05$ ).

Table 4: Haematological indices of broilers fed diets containing *Annona muricata* leaf meal.

Parameters	T1	T2	T3	T4	SEM	P-value
Wbc	167.10	169.13	157.80	150.73	3.58	0.25
Haemoglobin	11.63	11.27	11.20	11.43	0.24	0.94
Rbc	3.27	2.84	2.72	3.01	0.13	0.56
Mcv	137.23	139.90	136.73	136.23	0.66	0.21
Mch	45.73	46.17	43.13	47.37	0.67	0.11
Mchc	34.73	34.13	34.07	39.07	1.15	0.39
Plt	72.33	59.67	58.00	70.00	2.77	0.15
Pcv	34.57	33.60	33.57	33.60	0.71	0.96

WBC - total white blood cell count, RBC – red blood cell count, MCV – mean cell volume, MCH – mean cell haemoglobin estimation, MCHC – mean cell haemoglobin concentration, PLT – platelet count, PVC – packed cell volume

Growth performance of broilers fed diets containing *Annona muricata* leaf meal.

The growth performance of broilers fed diets containing AMLM is presented in table 5. The results show that the growth performance parameters were significantly different ( $P < 0.05$ ) except in initial weight which was not significantly different ( $P > 0.05$ ). The final weight of the broilers decreased as the quantity of AMLM increased. The average daily weight gain of treatment four was also lower than the other treatments. Average daily feed intake of the broilers also decreased as the quantity of AMLM increased while the feed conversion ratio of the broilers increased as the quantity of the *Annona muricata* leaf meal increased.

Table 5: Growth performance of broilers fed diets containing *Annona muricata* leaf meal.

Parameters	T1	T2	T3	T4	SEM	p-value
Inwt	0.06	0.05	0.05	0.06	0.001	0.330
Finalwt	2.81 <sub>b</sub>	2.78 <sub>b</sub>	2.55 <sup>a</sup>	2.36 <sub>a</sub>	0.062	0.003
Adwg	0.05 <sub>b</sub>	0.05 <sub>b</sub>	0.05 <sub>b</sub>	0.04 <sub>a</sub>	0.001	0.004
Totalfi	3.87 <sub>b</sub>	3.73 <sub>a</sub>	3.72 <sup>a</sup>	3.69 <sub>a</sub>	0.023	0.004
Adfi	0.55 <sub>b</sub>	0.53 <sub>a</sub>	0.53 <sup>a</sup>	0.53 <sub>a</sub>	0.003	0.010
Fcr	1.37 <sub>a</sub>	1.34 <sub>a</sub>	1.46 <sup>a</sup>	1.57 <sub>b</sub>	0.030	0.012

Final wt - Final weight, Inwt - Initial weight, Adwg - Average daily weight gain, Total fi - Total feed intake, Adfi - Average daily feed intake, Fcr - Feed conversion ratio

## V. DISCUSSION, CONCLUSION AND RECOMMENDATION

### Discussion

Haematological indices of broilers fed diets containing *Annona muricata* leaf meal.

The hematological indices of the experimental broilers fed diets containing varying levels of AMLM were not significantly different ( $P > 0.05$ ). The results obtained in this experiment agrees with the findings of Kuka et al (2024) who also reported no significant difference ( $p > 0.05$ ) in the hematological parameters of noiler birds injected *Annona muricata* leaf extract. Asa et al (2022) and Oke et al (2022) in contrast to the results obtained in this experiment recorded no significance differences ( $p > 0.05$ ) in all the hematological parameters except in RBC which was significant. The significant variations in the hematological parameters may be attributed to the age of the leaf meal and the concentration of the active ingredient in *Annona muricata* leaf meal. The values of the hematological indices obtained in this study were within the accepted range for broiler

chicken except for the WBC which was higher than the normal range. The levels of WBC of the broilers fed the treatment diets may be as a result of the presence of AMLM in the diets. Haemoglobin (Hb) concentration values obtained in this study was within the accepted range of 7.0-13.0(g/dl) for broiler chickens (Anon, 1980; Swenson, 1999). According to Oluwayinka et al (2021), leaf extract of *Annona muricata* reduced immunity response to Newcastle disease vaccine at 200mg/kg dosage but did not affect the haematology and the liver functioning of birds fed doses as high as 200mg/kg.

Growth performance of broilers fed diets containing *Annona muricata* leaf meal.

The results in Table 4.2 showed that birds fed diet with 0% AMLM gained significantly ( $P < 0.05$ ) higher weight than birds fed AMLM based diets. Birds on T2, T3, and T4 were observed to be statistically the same in average daily weight gain but its value decreased as inclusion of AMLM increased. This confirmed the observations made by Ash and Petaia (1992) and (Olugbemi et al., 2010) that increasing inclusion level of leaf meals in broiler diets results in decrease in growth performance. This observation could be generally traced to increasing fiber content of the diet which may have impaired nutrient digestibility and absorption (Ige et al., 2006) and Onu, (2010). It could also be attributed to the crude protein content or palatability of the control feed which enhances its acceptability and utilization. The negative effect of the anti-nutritional factors and phytochemical compounds present in AMLM on the birds could be responsible for decreased growth performance. There was also significant difference ( $p < 0.05$ ) in the average daily feed intake of the broilers. Average daily feed intake of the control diet was significantly higher than the average daily feed intake of the other treatment diets. According to Onunkwo and George (2015), leaf meals are generally bitter in taste, therefore, the inclusion of AMLM in the diets could have resulted in reduced palatability and thus reduce feed intake of the broiler diets. Omekam (1994) observed that unpalatable nature of a feedstuff will consequently prevent chicks from consuming adequate quantity of the feed. There was a significant increase in the feed conversion ratio of the birds fed AMLM -based diets than birds that were fed diets without AMLM. This suggests that

birds fed AMLM -based diets had poor utilization potential of the nutrients probably because of the increased bulkiness as inclusion level increased.

#### Conclusion and Recommendation

The results showed decreased growth performance and poor hematological indices of broiler fed diets containing AMLM at 2, 4 and 6% levels of inclusion. Therefore, lower levels of inclusion of AMLM should be experimented.

#### REFERENCES

- [1] Anon, I (1980). *Microbiology of Foods*, Vol 1. Factors affecting life and death of microorganisms. New York: Academic press.
- [2] Artawiguna, I. K. N., Nuriyasa, I. M., & Siti, N. W. (2023). Administration of fermented soursop (*Annona muricata*) leaf water extract through drinking water on the performance of joper male chickens. *International Journal of Life Sciences*, 7(2), 40–47. <https://doi.org/10.53730/ijls.v7n2.14403>.
- [3] Asa MN, Chamani M, Mousavi SN, Sadeghi A, and Foroudi F (2022). The effect of the in ovo injection of some carbohydrates and antioxidants on incubating parameters, blood and immunological parameters, intestinal morphometry, and post-hatching production performance in broiler chickens. *Italian Journal of Animal Science*, 21(1): 749-763. DOI: <https://www.doi.org/10.1080/1828051X.2021.1993092>
- [4] Ash A.J and Petaia L.A (1992), Nutritional value of *Sebania grandiflora* leaves for ruminant and monogastric. *Journal of Tropical Agriculture (Trinidad)* 69223-228.
- [5] Boluwaji O., Taniayo F. A., Taiye J. A., Elizabeth O. E., Angela O. and Miriam A (2024). Analysis of consumption expenditure on animal protein among farming households in Osun state, Nigeria.
- [6] Campbell, T.W. (1995) *Avian Hematology and Cytology*. 2nd Edition, Iowa State University Press, Ames.
- [7] Duncan, D.B. (1955) Multiple Range and Multiple F-Test. *Biometrics*, 11, 1-5.

- [8] FAO (2016). Experts' Network: Strengthening quality control systems in animal feed analysis laboratory in developing countries. Pp 1-7.
- [9] Ige A.O, Odunsi A.A, Akinlade J.A, Ojedapo L.O, Ameen S.A, Aderinola O.A and Rafiu T.A (2006), Gliricidia Leaf Meal in Layer's Diet: Effect on Performance Nutrient Digestibility and Economy of Production *Journal of Animal and Veterinary Advances* 5(6) 483 – 486.
- [10] Jiwuba Peter-Damian Chukwunomso, Kuka, Tar Timorthy1, Kadurumba, Ogechi Evangeline.2 Nwachukwu, Cynthia Ozioma (2017). " Evaluation of Haematological and Serum Biochemical Characteristics of *Annona muricata* (Sour Sop) Leaf Extract on Broiler Birds ", *International Journal of Research studies in Microbiology and Biotechnology*, vol. 3, no. 4, p. 6-11, 2017. <http://dx.doi.org/10.20431/2454-9428.0304003>
- [11] Kuka TT, N'nanle O, Karou S, Tona K, and Bakoma B (2024). Effects of in ovo Injection of Soursop (*Annona muricata*) Leaf Extract on Blood Profile, Immune Organs, and Intestinal Morphology of Noiler Chicks. *J. World Poult. Res.*, 14(4): 343-350. DOI: <https://dx.doi.org/10.36380/jwpr.2024.35>
- [12] Maertens L and Štruklec M. (2006). Technical note: Preliminary results with a tannin extract on the performance and mortality of growing rabbits in an enteropathy infected environment. *World Rabbit Sci.* 14:189-192.
- [13] Mohammed, A.K., Mohammed, G. and Akerejola, O.O. (2007). Haematological and serum biochemical changes in Bunaji work bulls after farmland ridging exercise in Kaduna State, Nigeria. *Journal of Animal and Veterinary Advances*, 6: 576-579.
- [14] Oke OE, Oyelola OB, Iyasere OS, Njoku CP, Oso AO, Oso OM, Fatoki ST, Bankole KO, Jimoh IO, and Sybill NI (2021). In ovo injection of black cumin (*Nigella sativa*) extract on hatching and post-hatch performance of thermally challenged broiler chickens during incubation. *Poultry Science*, 100: 100831. DOI: <https://www.doi.org/10.1016/j.psj.2020.10.072>
- [15] Olugbemi TS, Mutayoba S.K and Lekule F.P (2010), Effect of Moringa (*Moringa oleifera*) inclusion in cassava based diets fed to broiler chickens. *International Journal of Poultry Science* 9(4) 363 367.
- [16] Oluwayinka, E. B., Okunuga, F. S., Oni, O. O., Olukunle, J. O., Akinkuotu, O. A., & Oluwayinka, O. G. (2021). Activities of aqueous extracts of soursop (*Annona muricata*) leaves on immune response, haematology and serum biochemistry of broiler chickens. *Nigerian Journal of Animal Production*, 44(3), 110–116. <https://doi.org/10.51791/njap.v44i3.744>.
- [17] Omekam VN (1994), Studies on nutrition and health implication of dietary inclusion of dried poultry waste for broiler. MSc Thesis, Federal University of Technology; Owerri, Nigeria 41.
- [18] Onu P.N (2010), Impact of heat treated sheep manure-based diets with or without exogenous enzyme on nutrient digestibility and economics of production of finisher broilers. *International Journal of Science and Nature*.
- [19] Onunkwo D.N and George O.S (2015), Effects of Moringa *Oleifera* Leaf Meal on the Growth Performance and Carcass Characteristics of Broiler Birds.
- [20] Ritchie, J. and Spencer, L. (1994) Qualitative Data Analysis for Applied Policy Research. In: Bryman, A. and Burgess, R., Eds., *Anal. Qual. Data*, Routledge, London, 173-194. [https://doi.org/10.4324/9780203413081\\_chapter\\_9](https://doi.org/10.4324/9780203413081_chapter_9)
- [21] Russell, R.S. and Taylor, B.W. (1998) *Operations Management: Focusing on Quality and Competitiveness*. 2nd Edition, Prentice Hall, Inc., Upper Saddle River.
- [22] Suranto, A. (2011). The power of soursop against disease. Pustaka Bunda, Jakarta. Taylor, RSL and GHN Towers. 1998. Antibacterial constituents of the Nepalese herb, Centipeda minima. *Phytochemistry*. 47:631-634.
- [23] Swenson R. (1999). Differential diagnosis: a reasonable clinical approach. *Neurol Clin.* 1999 Feb;17(1):43-63. doi: 10.1016/s0733-8619(05)70113-4. PMID: 9855670.