

Haematology and carcass visual appraisal of broilers fed supplemental diets of *Aspilia africana*, *Azadirachta indica* and *Centrosema pubescence* leaf meals in Humid Tropical Nigeria

ABSTRACT

A study to determine the haematology and carcass visual appraisal of broilers fed basal feeds supplemented with different leaf meals was conducted. Four treatments: basal proprietary broiler feed only (T₁ - PBF) as control, basal proprietary broiler feeds with *Centrosema pubescence* (T₂ - PBF + CLM), *Azadirachta indica* (T₃ - PBF + NLM) and *Aspilia africana* (T₄ - PBF + ASLM) respectively, were utilized in a completely randomized design (CRD). On the last day of a 63-day feeding and growth trial, a set of 2ml blood samples were taken from 3 broilers per treatment into plastic tubes containing the anti-coagulant ethylene diamine tetra acetic acid (EDTA) for the determination of haematological parameters: PCV, Hb, RBC and WBC. The MCHC, MCH and MCV were also determined. Visual appraisals of their external body parts per treatment were also carried out. Results on the blood parameters of broilers fed leaf meals showed normal blood values recommended for healthy birds. This confirms that the use of leaf meals at 5% inclusion levels is not deleterious. Similarly, broilers fed with leaf meals showed a better appeal for their carcasses because of the yellow pigmentation of their body parts (shank, skin, beak and ear lobes). This will be an advantage to the consumers because it supplies vitamin A necessary for better vision. It is therefore recommended that poultry farmers include leaf meals in broiler feeds because it is not deleterious and can be of additional advantage due to the attractiveness of their carcasses to consumers.

Key words: Broilers, leaf meals, haematology, carcass, body parts, and visual appraisal.

1. INTRODUCTION

The feed crisis facing the poultry industry in Nigeria strongly suggests the need to investigate and utilize cheap and easily obtainable non-conventional feed resources. One of such non-conventional feed resources is leaf meal. The incorporation of protein from leaf sources in diets for broilers is fast gaining grounds because of its availability, abundance and relatively reduced cost. It had earlier been observed that leaf meal do not only serve as protein sources but also provide some necessary vitamins, minerals and also oxy-carotenoids which causes yellow colour of broiler skin, shank and egg yolk [1].

Comment [S1]: Check the right spelling and correct in the whole text. Is it *Aspilia* or *Aspilina*?

Comment [S2]: Could be used as a portion of conclusion

Comment [S3]: It was concluded

Many feed materials are fed to animals usually without recourse to their health and physiological implications on the animals. The commonest ~~parameter~~method for measuring these implications is through the haematology of the animals [2]. The comparison of an animal's haematological and biochemical values with a reference interval will provide evidence for numerous conditions such as infection, malnutrition and stress[3]. Furthermore, with the growing knowledge of the quality of animal protein supplies in human diets, and the preference for carcasses of good visual appraisal, it becomes imperative for poultry feed manufacturers and farmers to source tangible alternatives to cope with the current challenges in the meat market.

Plant leaves such as *Aspillia Africana*, *Azadirachta indica* (Neem) and *Centrosema pubescence* abound within the humid tropical environment of the South-south geopolitical zone of Nigeria and has not been maximally utilized as leaf meal supplements in poultry feeds. However, investigations on the use of the leaves especially *Aspillia Africana* and *Azadirachta indica* (Neem) has been on their utilization as traditional medicines in maintaining the health and welfare of both rural and urban dwellers in developing countries. There is therefore the need to investigate the effects of predominant plant materials: *Aspillia Africana*, *Azadirachta indica*(Neem) and *Centrosema pubescence* as leaf meal supplements on the haematology and ~~physical~~carcass appearance of broiler chickens in the area.

2. MATERIALS AND METHODS

2.1 Experimental site

The experiment was conducted at the poultry unit of the Teaching and Research Farm, Ignatius Ajuru University of Education Ndele Campus, Rivers State (Latitude 4⁰ 58' N and Longitude 6⁰ 48' E), Nigeria.

2.2 Processing of test ingredients

64 Fresh leaves from the young stems of the test ingredients: *Centrosema pubescence*,
65 *Aspillia Africana* and *Azadirachta indica* (Neem) were manually harvested from the bush and
66 fallow sections within the premises of the Ignatius Ajuru University of Education at Ndele
67 Campus. The fresh leaves were collected in batches in labeled bags. The leaves together with
68 their stems were spread on black polythene on an open floor in a greenhouse and air dried for
69 seven (7) days until it became crispy and shredded to separate the leaves from the stems. Air
70 drying of the leaves was carried out to reduce moisture content, to prevent fungal growth and
71 easy milling of the materials. The milling was done with a hand grinding machine, ~~operated~~
72 manually. The milled products represent the test materials which were incorporated
73 differently in the proprietary basal concentrate feeds.

74

75 2.3 Birds and distribution to treatment groups

76 A total of one hundred and eighty (180) day old of Marshal breed of broiler chicks
77 were brooded in the brooding unit (deep litter system) for 4 weeks (28 days) using stove and
78 kerosene lanterns. The chicks were randomly allotted to four (4) dietary feed treatment
79 groups: basal proprietary broiler feed only (T_1 - PBF) as control, basal proprietary broiler
80 feeds supplemented with *Centrosema pubescence* (T_2 - PBF + CLM), *Azadirachta indica*
81 (Neem) (T_3 - PBF + NLM) and *Aspillia africana* (T_4 - PBF + ASLM). The respective leaf
82 meals were incorporated at 5% supplemental rates into the basal proprietary broiler feeds.

83 2.4 Determination of Haematological Parameters

84 On the last day of a 63-day (9 weeks) experimental feeding and growth trial, a set of
85 blood samples were taken from 3 birds per dietary treatment. This was done by carefully
86 cutting the jugular veins of ~~the bird~~each bird and collecting 2ml samples ~~each~~ of blood into

87 | plastic tubes containing the anti-coagulant ethylene diamine_tetra_acetic acid (EDTA) for the
88 | determination of haematological parameters.

89 | The Haematological values of the blood samples were estimated for packed cell
90 | volume (PCV) and haemoglobin (Hb) concentration. Red blood cell (RBC) and total white
91 | blood cell (WBC) as well as the differential WBC counts were determined using the
92 | Neubauer_haemocytometer after appropriate dilution. Values for the constants: mean
93 | corpuscular haemoglobin concentration (MCHC), mean corpuscular haemoglobin (MCH)
94 | and mean corpuscular volume (MCV) were calculated from RBC, Hb and PCV values.

95

96 | **2.5 Visual appraisal on pigmentation of external body parts**

97 | At the end of the experiment after 63-days (9 weeks), the broilers from the leaf meal
98 | treatment groups (T₂, T₃ and T₄) and control (T₁ – without leaf meal treatment) were also
99 | physically assessed based on the yellowish pigmentation of the skin, beak, earlobe and shank.

100

101 | **2.6 Statistical Analysis**

102 | The haematological data obtained from birds in each treatment group were compared
103 | statistically on the basis of the different dietary treatments using Analysis of Variance
104 | (ANOVA) procedure for a Completely Randomized Design (CRD) [4]. Significant means
105 | were separated using the Duncan's New Multiple Range Test [4].

106

107 | **3. RESULTS AND DISCUSSION**

108

109 | **3.1 Nutrient composition of leaf meals and basal concentrate broiler finisher feeds**

The nutrient composition of the different leaf meals, the proprietary broiler feed (finisher) and those supplemented with the different leaf meals fed to the experimental broiler chickens is similar to those reported by [5].

3.2 Haematological Characteristics of broilers

Table 1 shows results of the haematological characteristics of broiler chickens exposed to the experimental diets: T₁- non- supplementation with leaf meal) used as control and other feeds supplemented with different leaf meals: T₂ (proprietary finisher basal feed supplemented with *Centrosema* leaf meal – PFBF + CLM), T₃ (proprietary finisher basal feed supplemented with Neem leaf meal - PFBF + NLM) and T₄ (proprietary finisher basal feed supplemented with *Aspillia* leaf meal - PFBF + ASLM) for 5 weeks after brooding (finisher phase).

The packed cell volume (PCV) value of the broilers fed with the experimental diets T₁ (leaf meal un-supplemented feed) and those supplemented with different leaf meal: T₂ (proprietary finisher basal feed supplemented with *Centrosema* leaf meal – PFBF + CLM), T₃ (proprietary finisher basal feed supplemented with Neem leaf meal - PFBF + NLM) and T₄ (proprietary finisher basal feed supplemented with *Aspillia* leaf meal - PFBF + ASLM) ~~are~~ were not significantly ($P > 0.05$) different. The PCV value of the broilers ranged from 31.00 – 33.33% for T₂ and T₁ respectively. The PCV values reported in the study for the broiler chickens were within the normal range (22 – 35%) reported by [6] for normal or healthy chickens. Similarly the values of PCV observed in this study for birds whose feeds were supplemented with Neem leaf meal (T₃) is within the range (30 – 40%) for broilers as reported by [1]. This implies that supplementation of broiler feeds with the different leaf meals has no deleterious effect on the broilers as they maintain their normal blood count [7].

The haemoglobin (Hb) value of the broilers fed with the experimental diets T₁ (leaf meal un-supplemented feed) and those supplemented with different leaf meal: T₂ (proprietary

finisher basal feed supplemented with *Centrosema* leaf meal – PFBF + CLM), T₃ (proprietary finisher basal feed supplemented with Neem leaf meal - PFBF + NLM) and T₄ (proprietary finisher basal feed supplemented with *Aspillia* leaf meal - PFBF + ASLM) ~~are~~were not significantly ($P > 0.05$) different. The haemoglobin value of the broilers ranged from 9.97 – 11.07g/dl for T₄ and T₁ respectively. The haemoglobin values reported in the study for the broiler chickens were within the normal range (7.0 – 13.0 g/d) reported by [6] for normal or healthy chickens. This implies that supplementation of broiler feeds with the different leaf meals has no deleterious effect on the broilers [7].

The Red blood cells (RBCs) value of the broilers fed with the experimental diets T₁ (leaf meal un-supplemented feed) and those supplemented with different leaf meal: T₂ (proprietary finisher basal feed supplemented with *Centrosema* leaf meal – PFBF + CLM), T₃ (proprietary finisher basal feed supplemented with Neem leaf meal - PFBF + NLM) and T₄ (proprietary finisher basal feed supplemented with *Aspillia* leaf meal - PFBF + ASLM) ~~are~~were not significantly ($P > 0.05$) different. The Red blood cells (RBCs) value of the broilers ranged from $3.40 - 3.70 \times 10^{12}/l$ for T₄ and T₁ respectively. The Red blood cells (RBCs) values reported in the study for the broiler chickens were within the normal range ($2.5 - 3.5 \times 10^{12}/l$) reported by [6] for normal or healthy chickens. This implies that supplementation of broiler feeds with the different leaf meals has no deleterious effect on the broilers as they maintain their normal blood count [7].

The Mean corpuscular haemoglobin concentration (MCHC) value of the broilers fed with the experimental diets T₁ (leaf meal un-supplemented feed) and those supplemented with different leaf meal: T₂ (proprietary finisher basal feed supplemented with *Centrosema* leaf meal – PFBF + CLM), T₃ (proprietary finisher basal feed supplemented with Neem leaf meal - PFBF + NLM) and T₄ (proprietary finisher basal feed supplemented with *Aspillia* leaf meal - PFBF + ASLM) ~~are~~were not significantly ($P > 0.05$) different. The Mean corpuscular

161 haemoglobin concentration (MCHC) value of the broilers ranged from 32.13 – 33.20g/100ml
162 for T₄ and T₂ respectively. The Red blood cells (RBCs) values reported in the study for the
163 broiler chickens were within the normal range (26.0 – 35.0 g/100ml) reported by [6] for
164 normal or healthy chickens. This implies that supplementation of broiler feeds with the
165 different leaf meals has no deleterious effect on the broilers as they maintain their normal
166 blood MCHC [7].

167 The Mean corpuscular haemoglobin (MCH) value of the broilers fed with the
168 experimental diets T₁ (leaf meal un-supplemented feed) and those supplemented with
169 different leaf meal: T₂ (proprietary finisher basal feed supplemented with *Centrosema* leaf
170 meal – PFBF + CLM), T₃ (proprietary finisher basal feed supplemented with Neem leaf meal
171 - PFBF + NLM) and T₄ (proprietary finisher basal feed supplemented with *Aspilia* leaf meal -
172 PFBF + ASLM) ~~are~~were not significantly ($P > 0.05$) different. The Mean corpuscular
173 haemoglobin (MCH) value of the broilers ranged from 28.47 – 30.83pg for T₄ and T₂
174 respectively. The Mean corpuscular haemoglobin (MCH) values reported in the study for the
175 broiler chickens were lower than the normal range (33.0 – 47.0 pg) reported by Banerjee,
176 (2004) for normal or healthy chickens. This variation may be due to difference in the breeds
177 of fowls [6].

178 The Mean corpuscular volume (MCV) value of the broilers fed with the experimental
179 diets T₁ (leaf meal un-supplemented feed) and those supplemented with different leaf meal:
180 T₂ (proprietary finisher basal feed supplemented with *Centrosema* leaf meal – PFBF + CLM),
181 T₃ (proprietary finisher basal feed supplemented with Neem leaf meal - PFBF + NLM) and
182 T₄ (proprietary finisher basal feed supplemented with *Aspilia* leaf meal - PFBF + ASLM)
183 ~~are~~were not significantly ($P > 0.05$) different. The Mean corpuscular volume (MCV) value of
184 the broilers ranged from 88.50 – 91.17fl for T₄ and T₂ respectively. However, although
185 broilers in the T₁ and T₂ exhibited higher MCV values, those of T₃ and T₁ were still within

Comment [S4]: Let references be uniform in the text

the normal values for birds. No mortality was recorded; hence the experimental diets were not deleterious.

188

Table 1: Haematology of broiler chickens fed basal concentrate diets supplemented with different leaf meals

Comment [S5]: Add P values of your ANOVA Tables

Parameters	Treatments				Mean	±SEM
	T1(PFBF)	T2 (PFBF + CLM)	T3(PFBF + NLM)	T4 (PFBF + ASLM)		
PCV (%)	33.33	31.00	33.00	30.33	31.92	2.88
Hb(g/dl)	11.07	10.50	10.97	9.97	10.63	0.85
RBC X10 ¹² /l	3.70	3.40	3.70	3.43	3.56	0.35
MCHC (g/100ml)	33.17	33.13	33.20	32.13	32.91	1.04
MCH (pg)	29.90	30.83	29.70	28.47	29.45	1.17
MCV (fl)	90.13	91.17	89.27	88.50	89.77	0.90

PFBF = Proprietary broiler Finisher Basal Feed; NLM = Neem Leaf Meal; CLM = Centrosema Leaf Meal; ASLM = Aspillia Leaf Meal; PCV = Packed cell volume; Hb = Haemoglobin; RBC = Red blood cell; MCHC = Mean corpuscular haemoglobin concentration; MCH = Mean corpuscular haemoglobin; MCV = Mean corpuscular volume.

195

3.3 Pigmentation pattern of external parts of broilers

Table 2 shows the pattern of pigmentation or colouration (white, fairly yellow or very yellow) of the external body parts (shank, skin, beak and ear lobe) of broiler chickens exposed to their basal feeds (non-supplemented/supplemented) with different leaf meals. Results from the visual appraisal revealed variations in the colour patterns of the treatment groups. It was observed that the leaf meal supplemented groups (T₂ = PFBF + CLM, T₃ = PFBF + NLM and T₄ = PFBF + ASLM) possessed yellowish pigmentations on their shanks, skin, beak and ear lobes. However, the degree of yellowness of these body parts slightly differed. Thus the T₂ (PFBF + CLM) treatment group showed very yellow (VY) colouration as compared to their fairly yellow (FY) (T₃ = PFBF + NLM and T₄ = PFBF + ASLM) counterparts. The body colouration of broilers in T₁ (PFBF only) showed white (W) colouration of the parts. The yellow pigmentation of the various leaf meal supplementation groups may be as a result of the presence some pigmenting agents in the different leaves.

209 This is can be attributed to the presence of several different xanthophylls of the general
 210 family of carotenoids resulting in pigmentation of the skin, fat, breast, and shanks and egg
 211 yolk [1].

212
 213 Table 2: Visual appraisal of pigmentation of body parts of broiler chickens fed basal
 214 concentrate diets supplemented with different leaf meals
 215

Treatment	Colour of external body parts											
	Shank			Skin			Beak			Ear lobe		
	W	FY	VY	W	FY	VY	W	FY	VY	W	FY	VY
T ₁ (PBFF only	Yes	No	No	Yes	No	No	Yes	No	No	Yes	No	No
T ₂ (PBFF + CLM	No	No	Yes	No	No	Yes	No	No	Yes	No	No	Yes
T ₃ (PBFF + NLM)	No	Yes	No	No	Yes	No	No	Yes	No	No	Yes	No
T ₄ (PBFF + ASLM)	No	Yes	No	No	Yes	No	No	Yes	No	No	Yes	No

216 PBFF = Proprietary broiler Finisher Basal Feed; NLM = Neem Leaf Meal; CLM =
 217 Centrosema Leaf Meal; ASLM = Aspilia Leaf Meal; W = White; FY = Fairly yellow; VY =
 218 Very yellow.
 219

220 4. CONCLUSION AND RECOMMENDATION

Comment [S6]: Too long

221
 222 The investigation on the blood parameters of broiler chickens fed leaf meal
 223 supplemented diets of Goat weed (*Aspillia Africana*), Neem (*Azadirachta_indica*) and
 224 *Centrosema pubescence* showed normal blood values as recommended for healthy birds. This
 225 further confirms that the use of the leaf meals at 5% inclusion level in basal broiler feeds is
 226 not deleterious to birds. Similarly, it was revealed that broilers whose feed were
 227 supplemented with different leaf meals showed a better appeal of their carcasses for
 228 consumption. The carcasses of the leaf meal treatment groups were better because of the
 229 yellow pigmentation of the different body parts (shank, skin, beak and ear lobes) due to the
 230 presence of carotenoids in these leaves. This will be of a better advantage to the consumers
 231 because of the supply of vitamin A that is good for better vision.

It is therefore recommended that poultry farmers resort to the inclusion of leaf meals in the feeds of broilers at 5% inclusion levels because this will not be deleterious and can be of additional advantage due to the attractiveness of their carcasses to consumers to improve their profit margins.

Comment [S7]: Already stated in the same sub-heading

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