Antioxidant Potentials of *Parquetina nigrescens* Leaf Extract Administration in Broiler Chicken Production

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ABSTRACT

This study looked into the grill chickens' *Parquetina nigrescens* (*P. nigrescens*) leaf extract (PNLE) antioxidant capacity. *P. nigrescens* leaves that were still fresh were collected from Ilishan-Remo in Ogun State, Nigeria. The experiment was carried out in the Babcock University Farmhouse's chicken department in Ilishan-Remo, Ogun State, Nigeria. After gathering the leaves and combining them with 50 g of the leaves in 1000 ml of water, the extract was produced. A total of 200-day-old Ross broiler chicks were divided into five groups at random and given different amounts of PNLE in 500 milliliters of water (0, 0.2, 0.4, 0.6, and 0.8 milliliters). The experiment lasted for 42 days. The design of the experiment was completely randomized design. Data was collected on performance, hematology, serum biochemical and antioxidant parameters, and temperature and humidity were monitored regularly. Data were subjected to a one-way analysis of variance ($P < 0.05$). There was no significant difference ($P > 0.05$) in performance characteristics. There was a significant difference ($P < 0.05$) in malonaldehyde with the 0ml PNLE group having the highest value (3.5 U/L X 10^9). There was a significant difference in catalase and superoxide dismutase with the 0.4 ml PNLE group having significantly highest ($P < 0.05$) values. Glutathione peroxidase was considerably ($P < 0.05$) impacted by PNLE administration. Total protein, albumin, globulin, glucose, cholesterol, triglycerides, urea, aspartate transaminase, and alanine transaminase were not substantially ($P > 0.05$) affected by PNLE delivery. The control group had considerably lower ($P < 0.05$) values for alanine phosphatase and red blood cell counts, and significantly higher ($P < 0.05$) values for creatinine. It is concluded that PNLE has good antioxidant potential thus improving the health status of broiler chickens.

Key Words: *Antioxidants, Oxidative stress, Parquetina nigrescens, Ross broilers*

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INTRODUCTION

Antioxidants are compounds that are known for their ability to neutralize free radicals, which are unstable molecules that can damage cells and contribute to various health problems. Antioxidants can be found in many foods, including fruits, vegetables, and whole grains, and they are also available in supplement form. In this essay, we will explore the different types of antioxidants and their potential health benefits [1].

Broiler chicken production plays a vital role in meeting the increasing global demand for poultry meat [2]. However, intensive farming practices and environmental stressors often lead to oxidative stress in broiler chickens,

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resulting in reduced productivity and compromised health [3, 4]. An imbalance between the body's antioxidant defense system and the generation of reactive oxygen species (ROS) leads to oxidative stress [5].

To mitigate the negative effects of oxidative stress, researchers have been exploring natural antioxidant compounds derived from various plant sources. *Parquetina nigrescens* (*P. nigrescens*) is a tropical plant that has been traditionally used for its medicinal properties. Many bioactive substances, such as tannins, flavonoids, and phenolic compounds, are known to be present in *P. nigrescens* leaves and have been linked to antioxidant activity [6].

Several studies have demonstrated the antioxidant potential of *P. nigrescens* leaf extract in various biological systems. In a study by [7], the authors evaluated the antioxidant properties of *P. nigrescens* leaf extract in rats exposed to oxidative stress. The results indicated that the extract exhibited significant antioxidant activity by scavenging free radicals and increasing the activities of antioxidant enzymes.

In a similar study conducted by [8], the antioxidant effects of *P. nigrescens* leaf extract were investigated in mice. The findings revealed that *P. nigrescens* contained antioxidants that can be valuable in attenuating reactions that generate free radicals in the body. Although previous research has shed light on the antioxidant properties of *P. nigrescens* leaf extract in other animal models, limited studies have focused specifically on its potential effects on broiler chicken production. Therefore, there is a need to investigate the impact of *P. nigrescens* leaf extract administration on broiler chickens to determine its suitability as a natural antioxidant supplement.

The current study aims to analyze the antioxidant potentials of *P. nigrescens* leaf extract administration in broiler chicken production. By examining various parameters such as antioxidant enzyme activities, lipid peroxidation levels, and immune responses, we aim to assess the potential benefits of incorporating *P. nigrescens* leaf extract as a natural antioxidant supplement in broiler chicken diets.

Understanding the antioxidant properties of *P. nigrescens* leaf extract and its effects on broiler chickens could provide valuable insights into sustainable strategies for enhancing poultry production and improving the overall health and well-being of these commercially important birds.

MATERIALS AND METHODS

Fresh *P. nigrescens* leaves were collected from Ilishan-Remo, Ikenne Local Government Area in Ogun State, Nigeria. This research was carried out at the Teaching and Research Farm, Babcock University, Ilishan-Remo, Ogun State, Nigeria. Ilishan-Remo is located in Nigeria's rainforest zone, with an annual rainfall of about 1500mm and a mean temperature of 27 degrees Celsius.

Five treatments were formulated. Treatment 1: 0ml of PNLE, Treatments 2, 3, 4, and 5 had 0.2, 0.4, 0.6, and 0.8 mil of *P. nigrescens* extract per 500ml of drinking water per bird administered respectively.

Experimental birds, management and design

Before the chicks arrived, the house was cleaned, disinfected, and let dry for fourteen days. The feeders and drinkers were completely cleaned and sanitized before the day-old chicks arrived. A commercial hatchery provided two hundred and eighty (200) Ross broiler chicks per day. The birds were weighed when they arrived and then randomly assigned to five treatments (T1, T2, T3, T4, and T5) in a completely randomized design consisting of four (4) repetitions, each with ten birds. For the duration of the 42-day study, the bird was given unlimited access to food and water. **Tables 1 and 2** showed the gross composition of the experimental diets and the proximate composition of the diets at both the starter and finisher phases.

Table 1. Gross composition of experimental starter and finisher diet (g/100 g)

Ingredient (kg)	Broiler starter	Broiler finisher
Maize	52.0	58.00
Soya bean meal	38.00	27.00
Wheat offal	4.59	9.64
Palm oil	2.00	2.00
Dicalcium phosphate	1.50	1.50
Oyster shell	1.00	1.00
Salt	0.25	0.25
Broiler premix	0.25	0.25
Methionine	0.30	0.25
Lysine	0.05	0.05
Avatec	0.06	0.06
TOTAL	100.00	1000.00
Calculated analysis		
Crude protein (%)	20.59	17.52
Crude fiber (%)	3.779	4.21
Metabolizable energy kcal kg	2,962.29	2,911.63
Determined Analysis		
Crude protein	20.50	18.00
Crude fibre	4.02	6.80

Table 2. Proximate composition of starter and finisher diets

Parameters (%)	Composition	
	Broiler starter	Broiler finisher
Moisture content	5.40 ± 0.20	5.50 ± 0.20
Crude protein	20.50 ± 0.40	18.00 ± 0.40

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Ether extract	3.00± 0.30	4.50 ± 0.30
Ash	10.50 ± 0.20	11.90 ± 0.20
Crude fibre	4.02 ± 0.30	5.80 ± 0.30

Data collection

Performance data, including feed intake, weight increase, feed conversion ratio (FCR), and blood chemical markers, were gathered (serum biochemistry, hematology, and antioxidant properties).

Feed intakes were calculated daily. This was done by deducting the amount of feed left in the feeders from the feed given on the previous day as feed intake for the day.

Feed intake (g) = feed given (g) – feed left (g) (1)

The weights of all the birds in each replicate were taken. This was done on the day of arrival and was subsequently done weekly until the end of the experiment.

Feed Conversion Ratio (FCR) = feed intake (g) ÷body weight (g) (2)

Hematology

On day 42, 2.0 ml of blood was collected from the wing vein (brachial vein) of birds (3 birds per replicate) into labeled sterile universal bottles containing Ethylene Diamine Tetra Acetate (EDTA) as an anticoagulant. The following hematological parameters were measured: Red blood cell count (RBC), white blood cell count (WBC), platelet, basophil, neutrophil, eosinophil, hemoglobin concentration, packed cell volume, monocytes, lymphocytes, heterophils according to the procedure of [9].

Serum biochemistry

At 42 d of age, the birds were fasted before blood collection but water was provided. 2.0 ml blood was collected from the wing vein of birds (3 birds per replicate) into heparinized bottles to determine serum biochemical components: Total Protein (TP), Globulin, Albumin, Urea, Creatinine, Aspartate aminotransferase (AST) and Alanine aminotransferase (ALT),

Anti-oxidative status

Birds were chosen at random when they were 42 days old, and blood samples were taken in heparinized tubes from the vein in their wing. Using commercial enzymatic kits, blood plasma levels of protein, glucose, triglycerides, and cholesterol were measured (Biosis LTD, Athens, Greece). The hydrophilic antioxidants in blood plasma were assessed using the oxygen radical absorbance (ORAC) assay to calculate the total antioxidant capacity (TAC).

The levels of creatinine, glutathione peroxidase, malondialdehyde, catalase, and superoxide dismutase were measured.

Statistical analysis

Data collected on performance, relative organ weight, and immunological response were subjected to analysis of variance (ANOVA) [10] and the treatment means were separated using the Duncan Multiple Range test where significant [11].

RESULTS AND DISCUSSION

Table 3 showed that there was no significant difference (P > 0.5) in the body weight, feed intake, and feed conversion ratio of the birds. Also, there was no significant difference (P > 0.05) in temperature and relative humidity.

Table 3. Performance and environmental indices of broiler chickens to administration of *P. nigrescens* leaf extracts in drinking water

	T1	T2	T3	T4	T5
Initial weight (g)	50.00 ± 0.00	52.00 ± 0.00	50.00 ± 0.00	51.00 ± 0.00	50.00 ± 0.00
Body weight (g)	1582.75 ± 79.86	1724.38 ± 59.68	1618.38 ± 43.44	1674.00 ± 65.28	1660.50 ± 30.93
Weight gain (g)	1532.75 ± 79.86	1672.38 ± 59.68	1568.38 ± 43.44	1623 ± 65.28	1616.50 ± 33.13
TFI/bird (g)	3313.68 ± 146.39	3552.33 ± 102.10	3063.10 ± 115.16	3237.39 ± 180.23	3470.25 ± 213.11
TWI/bird (ml)	835.00 ± 248.90	1761.72 ± 458.23	1882.98 ± 471.32	1911.30 ± 477.85	1915.62 ± 478.92
FCR	2.22 ± 0.20	2.15 ± 0.11	1.97 ± 0.11	2.02 ± 0.15	2.14 ± 0.11
Temp -morning (^{o}C)	26.26 ± 0.36	26.24 ± 0.47	28.15 ± 0.63	27.87 ± 1.12	26.65 ± 0.44
Temp-afternoon - (^{o}C)	33.09 ± 0.45	33.17 ± 0.21	32.89 ± 0.41	34.38 ± 1.31	33.07 ± 0.24
Temp - evening - (^{o}C)	28.05 ± 0.06	28.14 ± 0.09	27.94 ± 0.03	27.84 ± 0.63	26.88 ± 0.07
RH - morning (%)	54.38 ± 0.67	54.04 ± 0.79	55.21 ± 0.30	54.56 ± 0.66	53.46 ± 0.80
RH afternoon (%)	31.80 ± 0.11	31.98 ± 0.25	32.32 ± 0.30	31.86 ± 0.02	32.21 ± 0.12
RH-evening (%)	43.04 ± 0.85	43.86 ± 0.39	44.61 ± 0.37	43.89 ± 0.48	42.69 ± 0.38

*ab = Mean within the same row with different superscripts are significantly different.
Temp. – Temperature, RH – Relative Humidity

Table 4 shows the effect of the administration of *P. nigrescens* leaf extracts on the hematological response of the experimental birds. There was a significant difference ($P < 0.05$) in the red blood cell (RBC) count. The administration of *P. nigrescens* leaf extracts in drinking water did not significantly influence ($P > 0.05$) packed cell volume (PCV), hemoglobin (Hb), total white blood cell (TWBC) count, lymphocytes, platelets, monocytes, eosinophil, and basophils.

Table 4. Hematological responses of broiler chickens to administration of *P. nigrescens* leaf extracts in drinking water

PARAMETERS	T1	T2	T3	T4	T5
PCV (%)	33.00 ± 3.00	26.50 ± 1.50	30.50 ± 0.50	25.00 ± 3.00	28.50 ± 1.50
RBC COUNT (µ) * 10^6	3.60 ± 0.60^{a}	4.10 ± 0.80^{ab}	5.15 ± 0.35^{ab}	6.10 ± 0.50^{b}	5.05 ± 0.65^{ab}
TWBC (mm3)	23000.00 ± 2000.00	23000.00 ± 5000.00	19750.00 ± 2250.00	21750.00 ± 1750.00	20000.00 ± 2000.00
Hb(g/d)	11.00 ± 1.00	8.80 ± 0.50	10.15 ± 0.15	8.30 ± 1.00	9.50 ± 0.50
Platelet	175000.00 ± 25000.00	176500.00 ± 36500.00	157500.00 ± 12500.00	210000.00 ± 50000.00	259500.00 ± 29500.00
Heterophils	46.50 ± 7.50	45.00 ± 15.00	38.50 ± 5.50	45.00 ± 5.00	42.00 ± 8.00
Lymphocytes	40.00 ± 8.00	48.50 ± 16.50	47.50 ± 2.50	47.00 ± 7.00	48.00 ± 4.00
Monocytes	12.00 ± 1.00	5.00 ± 1.00	9.50 ± 2.50	6.00 ± 2.00	6.50 ± 3.50
Eosinophils	0.50 ± 0.01	1.00 ± 0.10	1.50 ± 0.50	1.00 ± 0.01	1.00 ± 0.01
Basophils	1.00 ± 0.01	0.50 ± 0.05	3.00 ± 0.0	1.00 ± 0.01	2.50 ± 0.50

Table 5 shows the serum biochemistry analysis of broiler chicken given *P. nigrescens* leaf extract at varying levels. There were no significant differences ($P > 0.05$) in the aspartate transaminase (AST), total protein (TP), albumin, glucose, globulin, alanine transaminase (ALT), and cholesterol, however, significant differences exist ($P < 0.05$) in alkaline phosphatase (ALP) and creatinine levels of the broiler chicken.

Table 5. Serum biochemical responses of broiler chicken administered varying dosages of *P. nigrescens* leaf extract

	T1	T2	T3	T4	T5
Glucose (mg/dl)	5.05 ± 0.65	4.85 ± 0.35	5.55 ± 0.05	5.10 ± 0.20	4.65 ± 0.05
Total Protein (mg/dl)	13.60 ± 0.10	13.45 ± 0.35	12.70 ± 0.20	12.90 ± 0.10	12.95 ± 0.35
Albumin (mg/dl)	9.15 ± 0.05	9.50 ± 0.70	8.40 ± 0.50	8.60 ± 0.40	8.75 ± 0.75
Globulin (mg/dl)	4.45 ± 0.05	3.95 ± 0.35	4.30 ± 0.70	4.30 ± 0.50	4.20 ± 0.40
AST(U/I)	70.00 ± 1.00	66.00 ± 1.00	66.00 ± 6.00	66.50 ± 3.50	74.00 ± 1.00
ALT(U/I)	90.00 ± 4.00	86.50 ± 4.50	95.50 ± 0.50	93.50 ± 3.50	94.50 ± 1.50
ALP (U/I)	29.50± 0.50^{ab}	31.00 ±3.00^{ab}	27.50 ± 4.50^{a}	38.00 ± 2.00^{b}	32.50 ± 0.50^{ab}
Creatinine (mg/dl)	0.17 ± 0.03^{b}	0.14 ± 0.01^{ab}	0.12 ± 0.01^{ab}	0.10 ± 0.01^{a}	0.15 ± 0.02^{ab}
Urea (mg/dl)	9.00 ± 1.00	10.00 ± 1.00	10.00 ± 2.00	8.00 ± 2.00	7.50 ± 0.50
Cholesterol (U/I)	29.50 ± 0.50	9.30 ± 0.30	11.00 ± 1.00	11.90 ± 2.10	10.90 ± 0.10
Triglyceride (U/I)	136.50 ± 13.50	143.00 ± 2.00	142.50 ± 3.50	149.00 ± 2.00	143.00 ± 5.00

^{ab} = Mean within the same row with different superscripts are significantly different, AST (Aspartate transaminase), ALT (Alanine transaminase), ALP (Alkaline phosphatase)

Table 6 presents the results of a study that investigated the effect of *P. nigrescens* leaf extracts on the antioxidant levels of broiler chickens. The study compared five treatment groups (T1-T5) and measured several biomarkers, including Malondialdehyde (MDA), Catalase (CAT), Superoxide dismutase (SOD), Total Antioxidant Capacity (TAC), and Glutathione Peroxidase (GPX). There were significant differences ($P < 0.05$) in malondialdehyde, catalase, superoxide dismutase, total antioxidants count, and glutathione peroxidase.

Table 6. Effect of *P. nigrescens* leaf extracts on the anti-oxidant properties of broiler chickens

	T1	T2	T3	T4	T5
Parameter	**0.0 ml/bird**	**0.2 ml/bird**	**0.4 ml/bird**	**0.6 ml/bird**	**0.8 ml/bird**
TAC (U/I)	10.40 ± 0.00^{c}	8.95 ± 0.05^{a}	12.25 ± 0.05^{e}	11.55 ± 0.05^{d}	9.60 ± 0.00^{b}
MDA (U/L X 10^{9})0	3.50 ± 0.01^{d}	3.24 ± 0.01^{c}	2.95 ± 0.01^{b}	2.94 ± 0.02^{b}	2.55 ± 0.01^{a}
CAT (U/I)	0.64 ± 0.01^{c}	1.17 ± 0.01^{d}	2.02 ± 0.02^{e}	0.51 ± 0.01^{b}	0.22 ± 0.01^{a}
SOD (U/I)	0.55 ± 0.05^{a}	0.45 ± 0.05^{a}	2.25 ± 0.05^{c}	1.00 ± 0.00^{b}	0.95 ± 0.05^{b}
GPx (U/I)	2.40 ± 0.00^{a}	5.00 ± 0.00^{e}	3.95 ± 0.05^{c}	3.75 ± 0.05^{b}	4.50 ± 0.00^{d}

P > 0.05: Significant difference observed ± standard error of the mean

*ab = Mean within the same row with different superscripts are significantly different

MDA – Malondialdehyde Concentration, CAT – Catalase, SOD – Superoxide dismutase, TAC – Total Antioxidants Counts, GPx- Glutathione Peroxidase, ALT – Alanine Transaminase, AST – Aspartic Transaminase.

There were no significant differences for all the performance characteristics measured. The results were in contrast with the report of Akintunde and Toye [12] who observed significant differences in weight gain, feed intake, and feed conversion ratio for Marshall broilers fed varying levels of *Moringa oleifera* seed meal. Also, Akintunde *et al.* [13] observed significant differences in body weight among Yoruba Ecotype Nigerian Local Chickens and Marshall broilers fed graded levels of *Moringa oleifera* seed meal. Also, the results were in contrast with the report of Akintunde *et al.* [14] who supplemented *Chromolaena odorata* leaf meal in the diets of broiler chickens. The results obtained from this study were also at variance with the reports of Oladele-Bukola *et al.* [15] who examined the growth and well-being of rabbits given solo or mixes of *P. nigrescens* and sunflower leaves. They found notable variations in the end weight, daily weight gain, total weight gain, and feed conversion ratio. The variations in results could be a result of the different test ingredients or the differences in the experimental animals used.

Hematological parameters have been recognized as one of the indicators for assessing the health status of animals [16]. According to Isaac *et al.* [17], animals with a healthy blood composition are probably going to perform well. There was no significant difference between the hematological parameters of birds administered *P. nigrescens* leaf extracts except the RBC count ($P < 0.05$). Red Blood Cells ($10^{\wedge 6}$ ul) value ranged between 3.60–6.10 and there was a significant difference in the values with birds with no administration of PNLE having the lowest values (3.60). The whole condition of chickens is influenced by their erythrocyte (RBC) count. Consequently, the test materials' (*P. nigrescens* leaf extracts) numerical improvements in red blood cell counts indicate that the blood's ability to carry oxygen was improved.

The administration of *P. nigrescens* leaf extracts did not have a significant effect on white blood cell differential counts. This was in contrast to the findings of Thachil and Bates [18], who observed that when an acute infection is present, the number of neutrophils in the blood increases quickly, and a blood count demonstrating this increase is important in the diagnosis of infections. The assertion that monocytes and basophils are often found in small to moderate quantities in the blood system was supported by the low values of these blood components.

It has been demonstrated that the amount and caliber of dietary protein affects both the total protein and creatinine contents [19-21]. The leaf extract's non-significant impact on the birds' total protein and albumin levels suggests that it can promote the synthesis of these blood components. However, the non-significant difference in the total serum protein observed among birds given *P. nigrescens* leaf extracts-based administration also suggests the nutritional adequacy of the diets and the safety of the test ingredient.

Although the intestinal mucosa, liver, bone, kidney, and placenta are the primary sources of alkaline phosphatase (ALP), intestinal ALP does not significantly raise serum ALP levels [22]. One blood enzyme that aids in the breakdown of proteins is called alkaline phosphatase (ALP). An ALP test measures how much ALP is circulating in the bloodstream. Having high or low ALP levels can indicate an underlying condition. Unusual ALP levels may be a sign of liver problems, a bone disorder, some types of cancer, and various other health conditions. Natural variations in ALP levels can happen even in the absence of underlying medical conditions. Nevertheless, aberrant levels may also indicate a serious illness, usually involving the kidneys, liver, or bones. Even though no specific inference could be made from the pattern of ALP values seen in the present study however, higher dosages of *P. nigrescens* leaf extracts could be belaboring the liver thus utilization of *P. nigrescens* leaf extracts at higher levels and longer duration beyond 42 days might pose future harm to the broiler birds.

There was a significant difference in the creatinine level and it had values ranging from 0.10 to 0.17 mg/dl with T1 (the control) having the highest and T4 (0.60 ml PNLE/500 ml water) having the lowest value. Because of

the animal's excess creatinine in the blood as a result of catabolism, creatinine is also associated with muscular atrophy [23]. The creatinine values observed in this study show that T1 (0ml/500ml of water) had the highest concentration of creatinine thus, the administration of *P. nigrescens* leaf extracts could be advantageous in combating excessive muscle wasting in broiler chickens.

There was a significant difference in the Malondialdehyde (MDA) with the control treatment (0ml PNLE) having the highest value. Malondialdehyde is considered a lipid peroxide. When its level increases, it can impair nucleic acid metabolism and function, destroy membrane proteins, and lead to autoimmune diseases [24, 25]. This implies that administering *Parquentina nigerescene* leaf extract at varying levels reduced significantly the MDA levels in the body of the birds.

The body uses enzymes like glutathione peroxidase and superoxide dismutase, which are vital to its defense against peroxidation, to secrete more of them to combat lipid peroxidation and harmful free radicals. In this investigation, the administration of Parquentina nigrescent leaf extract at higher doses markedly reduced MDA levels and elevated glutathione peroxidase levels.

Researchers found that the administration of *Moringa oleifera* leaf extracts significantly increased the levels of superoxide dismutase, catalase, and glutathione peroxidase in Wistar rats while decreasing the level of MDA [26-28]. Another study also reported that the administration of ginger extract significantly increased the levels of SOD and CAT, while decreasing the level of MDA in male rats [29-31]. There was a significant difference in catalase with birds administered 0.40 ml/500 ml of water exhibiting the best performance [32]. This shows that the rate of chemical reaction is greatly sped up by the administration of PNLE at 0.4 ml/500 ml water. There was a significant difference in Superoxide dismutase (SOD) with birds administered 0.4 ml PNLE having the highest value. This implied that the administration of *Parquentina nigerescens* leaf extract at 0.40 ml/500 ml water was able to effectively protect the cells from oxidative damage more effectively than the other levels of administration. There was a significant difference in glutathione peroxidase (GPx) with the birds in T2 (0.2 ml/500 ml water) having the highest values. This means that 0.2 ml of *Parquentina nigerescens* leaf extract shows it efficiently protected the cells from oxidative stress.

CONCLUSION

In conclusion, the study suggests that *Parquetina nigrescence* leaf extracts have antioxidant and anti-inflammatory properties in broiler chickens. The administration of *Parquetina nigrescence* leaf extracts in broiler chickens up to 0.80 ml/500 ml of water is hereby recommended for optimum health performance.

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