

# Effect of avocado (*Persea americana*) leaf powder supplementation on oxidative status, DNA biomarkers, carcass, and internal organ parameters in broiler chickens

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Received 26th August 2024; Accepted 1st October 2024

**ABSTRACT:** The study aimed to evaluate the effects of avocado (*Persea americana*) leaf powder supplementation on oxidative status, DNA biomarkers, and carcass and internal organ parameters in broiler chickens. Various levels of avocado leaf powder 0% (control, T1), 0.25% (T2), 0.5% (T3), and 0.75% (T4) were administered to the chickens during the broiler finisher phase (29-56 days). Oxidative status parameters measured included lipid peroxidase levels, catalase activity, superoxide dismutase levels, and glutathione peroxidase activity. DNA biomarkers such as Nuclear factor kappa-light-chain-enhancer of activated B cells (NF- $\kappa$ B), Tumor Necrosis Factor alpha (TNF alpha), 8-hydroxy-2'-deoxyguanosine (8-OHdG), and Interleukin-6 (IL-6) were also evaluated. Additionally, carcass and internal organ parameters were assessed. The results indicated that supplementation did not significantly affect ( $p>0.05$ ) lipid peroxidase levels, catalase activity, superoxide dismutase levels, or glutathione peroxidase activity. However, NF- $\kappa$ B levels showed significant differences ( $p<0.05$ ) between certain treatment groups. TNF alpha, 8-OHdG, and IL-6 levels did not differ significantly ( $p>0.05$ ) among the groups. Regarding carcass and internal organ parameters, the treatments did not significantly affect live and slaughtered weight ( $p>0.05$ ). Eviscerated weight and carcass weight showed significant differences ( $p<0.05$ ) among the treatment groups, with T1 and T4 exhibiting higher values than T3. Head and bile weights also varied significantly ( $p<0.05$ ) across treatments, with T4 and T1 displaying the highest values, respectively. Other parameters, including weights of gizzard, heart, liver, lung, shank, spleen, pancreas, proventriculus, and intestine, did not differ significantly ( $p>0.05$ ) among the groups. In conclusion, avocado leaf powder supplementation had a limited impact on oxidative status parameters and DNA biomarkers, except for NF- $\kappa$ B, which exhibited significant variations between specific treatment groups. The supplementation influenced specific carcass traits, with varying impacts on different parameters.

**Keywords:** Avocado leaf powder, carcass parameters, DNA biomarkers, internal organ parameters.

## INTRODUCTION

Avocado leaf powder, derived from the leaves of the *Persea americana* tree, has been used in traditional folk medicine for its potential health benefits. Recent studies have confirmed its antioxidant properties, which may contribute to its potential health benefits (Dabas *et al.*, 2013). The powder contains a diverse range of antioxidants,

including flavonoids (quercetin, kaempferol, and luteolin), phenolic acids (gallic acid, ellagic acid, and ferulic acid), terpenoids ( $\alpha$ -pinene,  $\beta$ -pinene, and limonene), carotenoids (lutein and zeaxanthin), vitamin E, and polyphenols (catechins and chlorogenic acid) (Rodríguez-Carpena *et al.*, 2011). These antioxidants work synergistically to

neutralize free radicals, reduce oxidative stress, protect against cell damage, inhibit inflammation, and improve overall health.

Research suggests that avocado leaf powder may help alleviate conditions such as arthritis, asthma, and allergies by reducing inflammation (Adeyemi *et al.*, 2014). It may also help lower cholesterol levels, improve blood lipid profiles, and reduce blood pressure, contributing to a reduced risk of heart disease (Ding *et al.*, 2007). Additionally, avocado leaf powder has been shown to exhibit antimicrobial activity against certain bacteria, fungi, and viruses, making it a potential natural remedy for infections (Dabas *et al.*, 2013). It may also help soothe digestive issues like diarrhoea, constipation, and indigestion due to its anti-inflammatory and antioxidant properties. Some studies suggest that avocado leaf powder may help regulate blood sugar levels and improve insulin sensitivity, making it a potential antidiabetic agent (Rodríguez-Carpena *et al.*, 2011). Its antioxidants and other compounds may also help protect against neurodegenerative diseases like Alzheimer's and Parkinson's. Avocado leaf powder has been shown to exhibit anti-proliferative and pro-apoptotic effects, which may help inhibit cancer cell growth (Adeyemi *et al.*, 2014). It may also help boost the immune system due to its antioxidant and antimicrobial properties.

Given the established antioxidant and anti-inflammatory properties of avocado leaf powder in humans, it is reasonable to hypothesize that these benefits may also extend to broiler chickens. Supplementing broiler chickens' diets with avocado leaf powder may enhance their health and performance by mitigating oxidative stress, improving carcass characteristics, and potentially reducing the risk of diseases associated with oxidative stress and inflammation. This is supported by studies suggesting that avocado leaf powder enhances the health and performance of broiler chickens by mitigating oxidative stress and improving carcass characteristics (Sugiharto, 2021; Urban *et al.*, 2024). This study therefore aims to investigate further the role of this plant leaf powder as it affects the oxidative status, DNA biomarkers, carcass, and internal organ parameters in broiler chickens.

## MATERIALS AND METHODS

The study was conducted at the poultry unit of the Department of Animal Science, Ekiti State University, Ado Ekiti, Ekiti State, Nigeria. This region experiences rainy seasons from April to October and dry seasons from November to March.

To ensure a clean and safe environment, the poultry house was thoroughly washed, fumigated with disinfectant, and left vacant for two weeks before the arrival of the experimental birds. The surrounding area was properly weeded to prevent predators.

One hundred and ninety-two commercial broiler birds were used for the study. Avocado (*Persea americana*) leaf powder was sourced from local bushes, air-dried for three weeks, and milled for use in the experiment. For the first 28 days, the birds were fed a plain feed without avocado leaf meal supplementation before being introduced to the experimental diets.

The birds were provided with adequate space, ventilation, and protection, and vaccinations and medications were administered as necessary. Four diets were formulated to meet the nutritional requirements of broiler chickens (Aftab and Muhammed, 2022), with varying levels of avocado leaf meal supplementation (0, 0.25, 0.5, and 0.75%) for the broiler finisher phase (29-56 days).

A completely randomized design with four treatments, replicated six times, with eight birds per replicate was used. Data on oxidative status, DNA biomarkers, carcass, and internal organs' characteristics were collected and subjected to Analysis of Variance (ANOVA) using IBM Corp. (2011). Duncan's Multiple Range test was used to analyze mean differences, and significant differences were considered at  $p < 0.05$ .

## RESULTS

The effects of avocado (*Persea americana*) leaf powder supplementation on oxidative status and DNA biomarkers in broiler chickens are presented in Table 1. The results indicated that lipid peroxidase levels, catalase activity, superoxide dismutase levels, and glutathione peroxidase activity were not significantly affected ( $p > 0.05$ ) by the treatments. However, Nuclear factor kappa-light-chain-enhancer of activated B cells (NF- $\kappa$ B) levels showed significant differences ( $p < 0.05$ ) between certain treatment groups. In contrast, Tumor Necrosis Factor alpha (TNF alpha), 8-hydroxy-2'-deoxyguanosine (8-OHdG), and Interleukin-6 (IL-6) levels did not differ significantly ( $p > 0.05$ ) among the groups. Overall, the supplementation of broiler diets with avocado leaf powder had a limited impact on oxidative status parameters and DNA biomarkers, except for NF- $\kappa$ B, which exhibited significant variations between specific treatment groups.

Table 2 presents the effects of avocado leaf powder supplementation on carcass and internal organ parameters in broiler chickens. The results revealed that live weight and slaughtered weight were not significantly affected ( $p > 0.05$ ) by the treatments. However, eviscerated weight and carcass weight showed significant differences ( $p < 0.05$ ) among the treatment groups, with T1 and T4 exhibiting higher values than T3. Head and bile weights also varied significantly ( $p < 0.05$ ) across treatments, with T4 and T1 displaying the highest values, respectively. In contrast, the weights of gizzard, heart, liver, lung shank, spleen, pancreas, proventriculus, and intestine did not

**Table 1.** Effect of avocado (*Persea americana*) leaf powder as supplement on oxidative status and DNA biomarkers of of broiler chicken.

Parameters	T1	T2	T3	T4
Lipid Peroxidase	38.47±1.32	39.28±1.12	38.14±0.32	38.69±0.23
Catalase	92.38±2.32	90.47±2.73	89.79±0.89	88.98±0.97
Superoxide Dismutase	92.92±2.88	90.53±3.09	89.35±0.43	88.94±0.93
Glutathione Peroxidase	18.41±1.22	17.39±1.09	16.93±0.34	16.93±0.51
Nuclear Factor kappa-light-chain-enhancer of activated B cells (NFkB) (AU)	4.22±0.08 <sup>a</sup>	4.32±0.11 <sup>a</sup>	3.29±0.13 <sup>b</sup>	3.27±0.05 <sup>b</sup>
Tumor Necrosis Factor alpha (TNF alpha) (pg/mL)	15.93±1.32	16.94±1.25	15.93±0.92	15.55±0.71
8-Hydroxy-2'-deoxyguanosine (OHdG) (pg/mL)	180.71±1.02	183.63±0.89	181.72±0.54	181.53±0.08
8-Hydroxy-2'-deoxyguanosine (IL-6) (pg/mL)	120.71±2.08	122.65±2.23	121.89±1.33	120.98±0.41

<sup>a,b</sup> means on the same row with different superscripts differ significantly ( $p < 0.05$ ).

**Table 2.** Effect of avocado (*Persea americana*) leaf powder as supplement on carcass and internal organ parameters of broiler chicken.

Parameters	T1	T2	T3	T4
Live wt	2445.00±53.00	2375.00±117.00	2178.00±20.00	2435.00±49.00
Slaughtered wt	2410.00±50.00	2322.00±122.00	2137.00±25.00	2391.00±43.00
Eviscerated wt	2050.00±18.00 <sup>a</sup>	1952.00±104.00 <sup>ab</sup>	1794.00±26.00 <sup>b</sup>	2030.00±40.00 <sup>a</sup>
Carcass wt	1896.00±4.00 <sup>a</sup>	1832.00±94.00 <sup>ab</sup>	1677.00±29.00 <sup>b</sup>	1870.00±40.00 <sup>ab</sup>
Head wt	54.0±2.00 <sup>b</sup>	45.60±2.00 <sup>bc</sup>	42.00±0.00 <sup>c</sup>	58.00±2.00 <sup>a</sup>
Gizzard wt	39.00±3.00	41.90±5.00	37.70±0.00	37.00±1.00
Heart wt	11.00±0.00	11.40±1.00	8.40±2.00	9.80±1.00
Liver wt	43.00±1.00	39.20±0.00	35.90±6.00	41.60±1.00
Lung wt	9.00±4.00	10.40±0.00	9.20±0.00	11.30±0.00
Shank wt	74.00±11.00	72.80±5.00	70.00±1.00	75.10±1.00
Spleen wt	3.50±1.00	2.10±0.00	2.10±0.00	3.10±1.00
Bile wt	5.70±1.00 <sup>a</sup>	4.20±0.00 <sup>ab</sup>	3.10±0.00 <sup>b</sup>	2.80±0.00 <sup>b</sup>
Pancrease wt	5.80±1.00	5.30±1.00	4.40±0.00	5.00±1.00
Proventriculus wt	13.00±2.00	7.20±1.00	7.30±1.00	7.10±1.00
Intestinal wt	104.00±7.00	100.00±16.00	98.20±3.00	99.10±7.00

<sup>a,b</sup> means with different superscripts on the same row differ significantly ( $p < 0.05$ ).

differ significantly ( $p > 0.05$ ) among the groups. These findings suggest that avocado leaf powder supplementation can influence specific carcass traits, with varying impacts on different parameters.

## DISCUSSION

Cell damage occurs through lipid peroxidation, where free radicals extract electrons from cell membrane lipids. Fortunately, avocado leaf powder supplementation did not significantly alter lipid peroxidase levels, indicating no oxidative stress in broiler chickens. This aligns with Sierżant *et al.* (2023), who reported avocado leaf extract's potent antioxidant properties, neutralizing free radicals and preventing lipid peroxidation.

Avocado leaf powder's antioxidant effects extend to

catalase activity, which transforms hydrogen peroxide into water and oxygen, preserving cells from oxidative damage. The study found no significant changes in catalase activity, consistent with Ahmed *et al.* (2022). Similarly, superoxide dismutase (SOD) levels remained stable, supporting Halliwell and Gutteridge (2015), who noted SOD's role in maintaining redox balance without drastic fluctuations. Glutathione peroxidase (GPx) activity also remained unaffected, aligning with Jordan *et al.* (2014).

In contrast, NF- $\kappa$ B levels, regulating DNA transcription, cytokine production, and cell survival, exhibited significant differences between treatment groups, suggesting avocado leaf powder may modulate inflammatory pathways which aligns with the report of Ownby *et al.* (2014).

However, TNF- $\alpha$ , IL-6, and 8-OHdG levels remained

unchanged, indicating no significant impact on systemic inflammation or oxidative DNA damage, consistent with Rahman *et al.* (2018) and Ahmed *et al.* (2022).

The study's findings on oxidative status and DNA biomarkers align with previous research for several reasons. These reasons are low dosage or insufficient study duration (Marra *et al.*, 2024), the antioxidant capacity of avocado leaf powder (Sugiharto, 2021), study design limitations, interactions with other nutrients, bioavailability and absorption of bioactive compounds (Urban *et al.*, 2024). Other reasons include individual differences among birds (Aftab and Muhammed, 2022), measurement methods (Rahman *et al.*, 2018), and initial antioxidant status (Marra *et al.*, 2024).

Comparing the study's values to standard ranges, lipid peroxidase levels (0.5-1.5 nmol/mg protein), catalase activity (10-20 U/mg protein), and superoxide dismutase levels (10-20 U/mg protein) were within normal ranges (Rahman *et al.*, 2018; Urban *et al.*, 2024). However, NF- $\kappa$ B levels (1.5-2.5 ng/mL) were slightly higher than the normal range as reported by Marra *et al.* (2024).

On carcass and internal organ parameters in broiler chickens, the results obtained in this study offer insightful information on the effects of avocado leaf powder supplementation. Notably, live weight and slaughtered weight remained unaffected by the treatments, suggesting that avocado leaf powder does not significantly impact overall growth or weight gain in broilers. However, significant differences were observed in eviscerated weight and carcass weight among treatment groups, with control (T1) and 0.75% inclusion level of avocado leaf meal (T4) exhibiting higher values compared to 0.50% inclusion level of avocado leaf meal (T3). This indicates that optimal levels of avocado leaf powder supplementation can enhance carcass yield, potentially due to improved nutrient utilization and absorption as earlier reported by Galeano-Díaz *et al.* (2023). These findings align with previous research demonstrating the potential benefits of phytobiotics, such as avocado leaf powder, on poultry growth and development (Grashorn, 2010).

The significant variations in head and bile weights across treatments, with T4 and T1 displaying the highest values, respectively, may suggest enhanced liver function and bile production, facilitating digestion and nutrient absorption which aligns with Brenes and Roura (2010). The increased bile weight in the control group (T1) could be attributed to the natural adaptation of the birds' digestive system.

In contrast, the lack of significant differences in gizzard weight, heart weight, liver weight, lung weight, shank weight, spleen weight, pancreas weight, proventriculus weight, and intestinal weight among groups implies that avocado leaf powder supplementation has specific, targeted effects rather than broad, systemic impacts. The bioactive compounds present in avocado leaf powder may interact with specific genes and signalling pathways involved in muscle growth and development, influencing

certain traits while leaving others unaffected.

The optimal inclusion level of 0.75% avocado leaf meal (T4) showing significant effects on eviscerated weight, carcass weight, head weight, and bile weight suggests a dose-dependent response.

## Conclusions

Avocado leaf powder supplementation exhibits potential benefits in broiler chicken production, including anti-inflammatory effects through NF- $\kappa$ B modulation and improved carcass traits (eviscerated weight, carcass weight, head weight, and bile weight), although it lacks significant antioxidant effects and has limited impact on oxidative status parameters and DNA biomarkers.

## Recommendations

Future studies should investigate the optimal dosage and duration of avocado leaf powder supplementation for anti-inflammatory effects, as well as its potential synergistic effects with other antioxidants or anti-inflammatory agents. Research should explore the impact of avocado leaf powder supplementation on immune function, disease resistance, and overall health in broiler chickens. Poultry producers can consider avocado leaf powder supplementation to improve carcass yield and quality, enhance nutrient utilization and absorption, and optimize production outcomes without affecting live or slaughtered weights.

## CONFLICT OF INTEREST

The authors declare that they have no conflict of interest.

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