

Article

Blood Parameters, Kidney Histology and Growth Performances in *Gallus gallus* Domesticus (Brahma) Hens Fed a Diet Supplemented with *Dacryodes edulis* (Safou) Powder Leaves

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Abstract: The leaf extracts of *Dacryodes edulis* possess high concentrations of alkaloids, saponins, flavonoids, and tannins with various biological activities, including antimicrobial, antifungal, and antioxidant activities. These activities can be used in animal production to avoid the energy lost in favor of growth and reproduction. A total of 48 Brahma hens (45 days old), weighing on average 400 ± 12 g, were randomly distributed into four dietary treatment groups (12 birds each) with four replicates per group. The control group (T0) received 0% *D. edulis*, while the three test groups (T0.25, T0.50, and T0.75) were given feed with *D. edulis* powder leaves at concentrations of 0.25, 0.5, and 0.75%, respectively, for a period of 60 days. Water and feed were supplied ad libitum. At the end of the study period (60 days), eight birds per treatment (two per replicate) were fasted, weighed, and slaughtered. Blood samples and organs were collected for analysis of growth characteristics, oxidative stress, and toxicity indices. This study revealed a significant ($p < 0.05$) increase in feed intake and live body weight with 0.75% *D. edulis* powder leaves. Abdominal fat was found to be significantly ($p < 0.05$) lower with 0.75% *D. edulis* powder leaves compared to the control group. Serum Aspartate aminotransferase activity was significantly ($p < 0.05$) lower in birds exposed to 0.75% *D. edulis* leaf powder compared to the control group. The use of *D. edulis* leaf powder as feed additive in feed could reduce oxidative stress and improve growth performance in Brahma. More research can be conducted on *D. edulis*, and it can be used in broiler feed at 0.75% concentration, which has shown a significant increase and decrease, respectively, in live body weight and serum aspartate aminotransferase activity.

Keywords: Brahma hens; *Dacryodes edulis* leaf powder; growth performance; oxidative stress; toxicity

1. Introduction

In poor and developing countries, the extensive or traditional poultry farming system is a means of subsistence for populations in urban and peri-urban areas [1,2]. According to Fotsa et al. [3], 80% of the poultry population in rural areas is made up of local breeds of chickens. In Cameroon, local chicken populations represent about 56% of the national poultry population [3], with high production in the Far North, West, and North West regions. The intensification of local poultry farming is linked to their adaptation to difficult village or rural conditions and their various interests. Traditional poultry farming plays a key role in the quest for self-sufficiency and sustainable food security. The village chicken plays an important socio-cultural role. It is used in wedding ceremonies, in traditional pharmacopoeia, and in maintaining social cohesion within rural communities [4]. In addition, traditional poultry represents a source of income for poor rural farmers, especially

women, and even for the state economy, as well as organic fertilizer for agriculture [5]. Despite their many advantages, village chickens have poor growth and reproductive performance compared to exotic strains. At adulthood (24 weeks to one year), the average live weight of a local hen varies from 900 g to 1.45 kg and that of a rooster from 1.3 to 2.4 kg [6,7]. In terms of meat production, carcass yields vary from 61 to 79%, with roosters generally having values 7–12 points higher than hens [1,6].

The low values of traditional poultry characteristics would be related to their genetic potential and the synergistic or non-synergistic effects of multiple environmental factors, such as poor water and feed quality and quantity, temperature fluctuations, exposure to various high-burden microbes, and inadequate housing associated with high densities around waterers and feeders [8].

In the search for solutions to reduce or neutralize the impact of multifactorial constraints related to the living environment of local hens, available and less harmful products with diversified biological activities, such as medicinal plant-based products, should animate the interest of stakeholders in the field. Medicinal plants, among which the safou (*Dacryodes edulis*), rich in various bioactive molecules capable of limiting the attacks of the various environmental factors, should be exploited.

Dacryodes edulis belongs to the family of Burseraceae and is a dioecious plant found natively in many African countries, including Cameroon [9]. It is a perennial tree crop, which is widely grown in the wet regions of West Africa and Cameroon [10]. Phytochemical studies of the leaf extracts of *Dacryodes edulis* revealed high concentrations of alkaloids, saponins, flavonoids, and tannins [9,11], and they show various biological activities, which include antimicrobial, antifungal, and antioxidant activities [9,11]. The leaf sap is instilled into the ear for ear problems, and a decoction of the leaves is prepared as vapour for feverish stiffness with headache [12]. It was reported that the leaves were made into plaster to treat snakebite in Southwest Cameroon [13]. Based on the diversity of bioactive compounds and biological activities of *Dacryodes edulis*, we believe that it could positively influence growth performance, oxidative stress, and toxicity indices in birds. This study seeks to ameliorate the production of an indigenous poultry breed using medicinal plants.

2. Materials and Methods

2.1. Study Area and Duration

The experimental work was carried out from October to December 2022, at the poultry unit of the Teaching and Research farm (FAR-UDs) of the Faculty of Agronomy and Agricultural Science of the University of Dschang, West region of Cameroon. The farm is situated at latitude 5°44'–5°36' and 5°44'–5°37' North and longitude 10°06'–9°94' and 10°06'–9°85' East and at an altitude of 1420 m above sea level. The climate of the region is equatorial, of the Cameroonian type, modified by altitude. Annual temperature varies between 10 °C (July–August) and 25 °C (February). Relative humidity varies between 40–97% and rainfall ranges from 1500 mm to 2000 mm per annum. The dry season goes from mid-November to mid-March and the rainy season from mid-March to mid-November, corresponding to the cultivation season [14].

2.2. Animal Material

A total number of 48 healthy Brahma hens, age 45 days old, weighing averagely 400 ± 12 g were purchased from COSEPVIM (Societe Co-Operative Des Eleveurs De Poulets Villageois De La Mifi) Bafoussam and were raised over a 60-day period. Throughout this period, the animals were provided with local formulated poultry feed and water ad libitum. The birds were sexed then identified with a ring bearing their number on one of its paws.

2.3. Housing

During the experiment, the animals were housed in one of the buildings of the Teaching and Research Farm (FAR-UDs) of the Faculty of Agronomy and Agricultural Science of the

University of Dschang. The birds were housed in galvanized metals wire cages sub divided into five compartments each, in which the birds were kept at room temperature under 12 h of light and 12 h of dark cycle. The birds were kept at a density of 0.12 m² per bird.

2.4. Feeding

The birds were provided with a local feed of 70 g per bird per day during the starter phase and 100 g of feed per bird per day during the grower phase. The animals received feed and fresh water ad libitum and the feed constitute of ingredients bought at the Dschang market. The feed was provided to the birds in plastic containers of 1.5 litter and water in 1 litter containers. Each bird was provided with feed mixed with *D. edulis* leaf powder at various percentages. The feed composition and the chemical characteristics at the starter and grower phase are presented on Table 1.

Table 1. Composition and chemical characteristics of experimental rations used (starter phase and grower phase).

Ingredients (%)	Starter Phase (1–12 Weeks)	Grower Phase (13–20 Weeks)
Maize	48.00	45.00
Wheat bran	2.50	0.00
Wheat middings	14.00	22.00
Cotton seed cake	8.00	6.00
Soya bean meal	15.00	13.00
Fish meal	6.00	3.00
Bone meal	0.00	1.50
Shell	1.50	1.00
Concentrate 5% (*)	5.00	5.00
Palm oil	0.00	3.50
Total	100.00	100.00
Calculated chemical composition		
Crude protein (%)	23.20	20.70
Metabolizable energy (kcal/kg)	2913.00	3013.00
Calcium (%)	1.48	1.51
Phosphorus (%)	0.69	0.73
Lysine (%)	1.29	1.10
Methionine (%)	0.43	0.40
ME/CP	125.00	145.00

* Broiler concentrate 5%: Crude protein = 40%; Metabolizable Energy = 2078 kcal/kg; Calcium = 8%; Available Phosphorus = 2.05%; Lysine = 3.30%; Methionine = 2.40%. ME: Metabolizable Energy; CP: Crude protein.

2.5. Sanitary Protection

Two weeks before arrival of chicks, the livestock building and various equipment were cleaned and disinfected using Javel and Cresyl solutions (20 mL per 1 litter of water), spread in the room and on all the cages. The floor of the room was equally disinfected by applying calcium carbonate on the walls and floor of the room. After disinfection, a crawl space of two weeks was allowed before introduction of the birds. The room and equipment used were cleaned on a daily basis. Upon arrival of the birds in the farm, they were provided with Aliseryl anti-stress (1 g per 2 litters of water) for three consecutive days during the period of adaptation.

2.6. Plant Material

Fresh matured leaves of Safou were harvested on the same tree at campus G of the University of Dschang during the month of February 2020 between 8–10 am. The *Dacryodes edulis* leaves were washed and shadow dried at an ambient temperature of 25 °C under a shade for ten days and milled into fine powder using a grinding machine. The

powder was then sieved to obtain fine Safou leaf powder, which was then sealed in an air-tight bag and stored in a cool dry place until it was used.

The phytochemical screening of Safou powder leaves was then carried out at the laboratory of chemistry of natural substances of the University of Dschang based on the method described by Wagner et al. [15] and Hussain et al. [16] in order to confirm the presence or absence of certain bioactive constituents, such as flavonoids, phenols, alkaloids, tannins, steroids, saponins, and terpenes. An amount of 50 g of the Safou leaves powder were used, and the result obtained was presented in Table 2.

Table 2. Test used and chemical constituent of *D. edulis* leaves powder.

Tests	Constituents	Safou Leaf Powder
Shinoda	Flavonoids	++
	Tannins	+
Chlorine fluoride	Phenols	++
Dragendorff	Alkaloids	—
Foam index	Saponins	++
Lieberman–buehard	Steroids	++
	Terpenoids	++

—: absent, +: weak, ++: high.

2.7. Trial Conduction and Experimental Design

In this study, 48 Brahma birds, with characteristics given above, were weighed at the beginning of the experiment and randomly assigned to four treatments groups (T0, T0.25, T0.50 and T0.75) with 12 birds per group. Before trial conduction, a period of adaptation of one week was observed to permit the birds to adapt themselves to the new environment. These birds were fed ad libitum from day one to the end of the experiment using the experimental diets. Birds in the control group were fed with a commercial diet without supplement. Meanwhile, birds of the other three test groups, during the same period, received diets supplemented with 0.25%, 0.50%, and 0.75% of *Dacryodes edulis* leaf powder, respectively.

T0 = Control (formulated feed without supplement)

T0.25 = Formulated feed + 0.25% of *D. edulis* leaf powder

T0.50 = Formulated feed + 0.5% of *D. edulis* leaf powder

T0.75 = Formulated feed + 0.75% of *D. edulis* leaf powder

2.8. Studied Parameters and Data Collection

The birds were weighed at the beginning of the trial and once a week throughout the trial. They were weighed using a balance of capacity of 7000 g and precision of 1 g for the determination of growth performance.

2.8.1. Growth Characteristics

Feed intake

Feed was weighed at the beginning of each week and distributed daily to the birds. Remains of each replicate of the experimental treatments were weighed daily with the aid of a balance of 1 g precision and capacity of 7 kg. Feed intake was then evaluated by making the difference between the quantity of feed served during a week and remains for the same week of that period.

Live body weight (LBW) and weekly body weight (WBWG)

The animals were weighed at the start of the trial and after every seven days subsequently between 6–8 am. Live body weight for individual birds were recorded weekly; weekly body weight gain was obtained by making the difference in live body weight of two consecutive weeks according to the procedures of Mc Donald et al. [17] ($WBWG = P_n - P_{n-1}$). This was performed with the aid of an electronic balance described earlier above. During the duration of the experimental period, the birds of each group were weighed individually.

$$\text{WBWG} = P_n - P_{n-1}$$

WBWG = Weekly body weight gain

P_n = Live body weight of the week considered

P_{n-1} = Live body weight of the preceding week

Daily weight gain (DWG)

Daily weight gain was obtained by dividing the weekly weight gain by seven.

Feed Conversion ratio (FCR)

Weekly feed conversion ratio was calculated by dividing the quantity of feed consumed weekly by the weight gain of the animals for the same week.

$$\text{FCR} = \frac{\text{Weekly feed intake (g)}}{\text{weekly weight gain (g)}}$$

Feed efficiency (FE)

FE was obtained by making the inverse of feed conversion ratio.

$$\text{FE} = \frac{1}{\text{Feed conversion ratio}}$$

Relative weight, volume and density of organs

At the end of the experiment (60th day), eight birds per treatment were randomly selected and fasted for 24 h, weighed, and slaughtered by decapitation. After slaughtering of the animals, the abdominal cavity was opened and organs, such as the liver, heart, and intestine of the various birds, were carefully removed free of all adipose tissues and weighed separately using a balance of capacity of 160 g and precision 10^{-3} g. The relative weight of each organ was calculated using the following formula:

$$\text{Relative organ weight (\%)} = \text{Organ weight (g)} / \text{Live body weight (g)} \times 100$$

The volume of liver was determined by the water displacement method that consisted of pouring a quantity of 0.9% NaCl in a graduated cylinder at an initial known volume and reading the volume (V_1). Then, the organ was introduced, and the volume (V_2) was noted. The volume of the organ was then calculated using the following formula:

$$\text{Vorgan} = V_2 - V_1$$

The density of the intestine was calculated using the following formula described by Abdel-Fattah et al. [18]:

$$\text{Density of intestine (g/cm)} = \frac{\text{Weight of intestine}}{\text{Length of intestine}}$$

2.8.2. Serum Biochemical Analysis

Blood Collection

At the end of the experiment (60th day), birds were fasted for 24 h and 2 birds from each replicate was selected and sacrificed by sectioning of the jugular vein. Approximately 4 mL of blood samples from each selected bird was collected after sectioning of the jugular vein into tubes free of anticoagulant for biochemical dosage. After clotting, the serum was separated from clotted blood by centrifugation at 3000 rpm for 15 min, then collected using a micro pipette and introduced in Eppendorf tubes. The collected serum was then stored at -20°C until use, as described by Mohamed and Nagwan [19].

Biochemical Markers

Part of the serum was used for the determination of aspartate aminotransferase (AST), alanine aminotransferase (ALT), creatinine, urea, and total protein contents using commer-

cial kits CHRONOLAB, Ref. 101-0255, CHRONOLAB, Ref. 101-0256, CHRONOLAB, Ref. 101-0281, CHRONOLAB, Ref. 101-0575, and CHRONOLAB, Ref. 101-0240, respectively, by spectrophotometer methods, as described by CHRONOLAB commercial kits.

2.8.3. Histological Studies

Realization and observation of histological sections of the kidney were performed in the laboratory of anatomo-pathology of Banjoun Evangelic University following a specific protocol. The same section of the kidney collected was fixed immediately in 10% formalin and processed for histopathological studies. Sections of 4 μ m thick were cut and stained using hematoxylin and eosin procedure, as described by Bancroft and Gamble [20], and they were examined microscopically. For each slide, a minimum of 20 microscopic field (20X)/slide was examined and evaluated.

2.8.4. Estimation of Oxidative Stress

Evaluations of malondialdehyde and activities of superoxide dismutase, total peroxidase, and catalase were used as markers of oxidative stress and tissue damage according to the methods described, respectively, by Nilsson [21], modified by Kodjio et al. [22], Sehrlirli et al. [23], Misra and Fridovich [24], and Sinha [25].

2.9. Statistical Analysis

The statistical analysis was carried out using the SPSS 22.0 software. Results were expressed as mean \pm standard deviation. Differences between groups were assessed using one way ANOVA followed by Duncan post hoc test with the significance level set at 0.05. p -value calculation was performed using the student t -test. A p value of less than 0.05 was considered as significant. The normality of data was tested by the Shapiro-Wilk test. The relationships between different parameters were highlighted by the correlation coefficient of Bravais-Pearson. The nonlinear regression was used to determine the relationship between growth characteristics and the concentration of *Dacryodes edulis* in feed.

The statistical model used was: $X_{ij} = \mu + \alpha_i + e_{ij}$,

X_{ij} : The observed value of each of the response variables;

μ : Overall population mean;

α_i : Observed effect of the dietary treatments;

e_{ij} : Residual or random error due to the experimentation.

3. Results and Discussion

3.1. Results

3.1.1. Effects of *D. edulis* Leaf Powder on Growth Characteristics of Brahma Hens

The effect of *Dacryodes edulis* powder leaves in feed on average feed intake, live body weight, weekly body weight gain, daily weight gain, feed conversion ratio (FCR), and feed efficiency are illustrated on Figures 1–4 and summarized in Table 3.

Table 3. Effects of Safou (*Dacryodes edulis*) leaves powder on growth characteristics in Brahma hens.

Growth Characteristics	Control T0	<i>D. edulis</i> Concentration (%)			p Value
		T0.25	T0.50	T0.75	
FI(g)	4692.92 \pm 252.35 ^{ab}	4439.08 \pm 494.23 ^b	4728.41 \pm 239.70 ^a	4823.64 \pm 38.58 ^a	0.03
LBW (g)	1330.00 \pm 153.14 ^b	1466.50 \pm 111.44 ^{ab}	1403.43 \pm 111.78 ^{ab}	1510.00 \pm 44.47 ^a	0.04
WBWG (g)	998.25 \pm 280.23	916.83 \pm 343.71	1027.67 \pm 209.13	1117.91 \pm 319.39	0.45
DWG (g)	16.64 \pm 4.67	15.28 \pm 5.73	17.13 \pm 3.48	18.63 \pm 5.32	0.45
FCR	5.60 \pm 3.78	5.79 \pm 3.14	4.84 \pm 1.38	4.70 \pm 1.53	0.73
FE	0.21 \pm 0.06	0.20 \pm 0.07	0.22 \pm 0.067	0.23 \pm 0.07	0.74

^{a,b}: values affected with the same letter in the same line are not significantly different ($p > 0.05$). FC: Feed intake; LBW: Live body weight; WBWG: Weekly body weight gain; DWG: Daily weight gain; FCR: Feed conversion ratio; FE: Feed efficiency; P: Probability; T0: Control (0% additive); T0.25: Control diet containing 0.25%; T0.50: Control diet + 0.5%; T0.75: Control diet + 0.75 of *D. edulis* leaves powder, respectively.

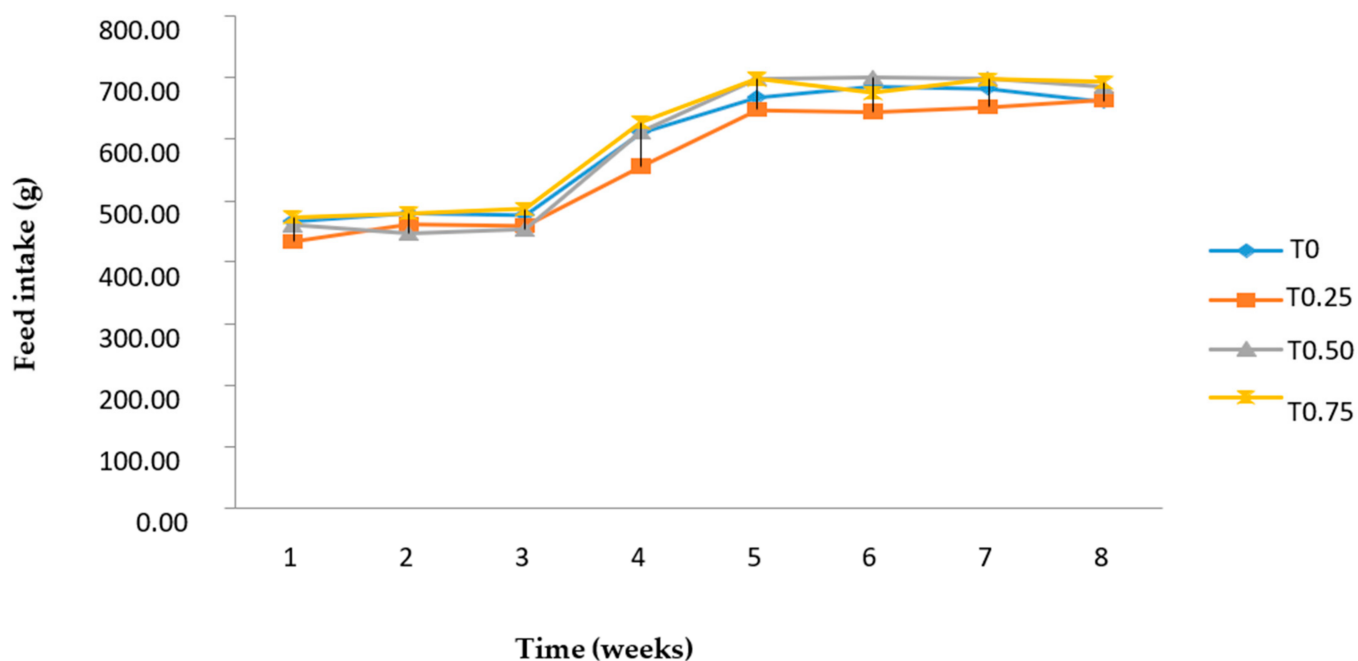


Figure 1. Weekly evolution of feed consumption as influenced by the rate of *D. edulis* leaves powder in feed of Brahma hens. T0: Control (0% additive); T0.25: Control diet containing 0.25%; T0.50: Control diet + 0.5%; T0.75: Control diet + 0.75 of *D. edulis* leaves powder, respectively.

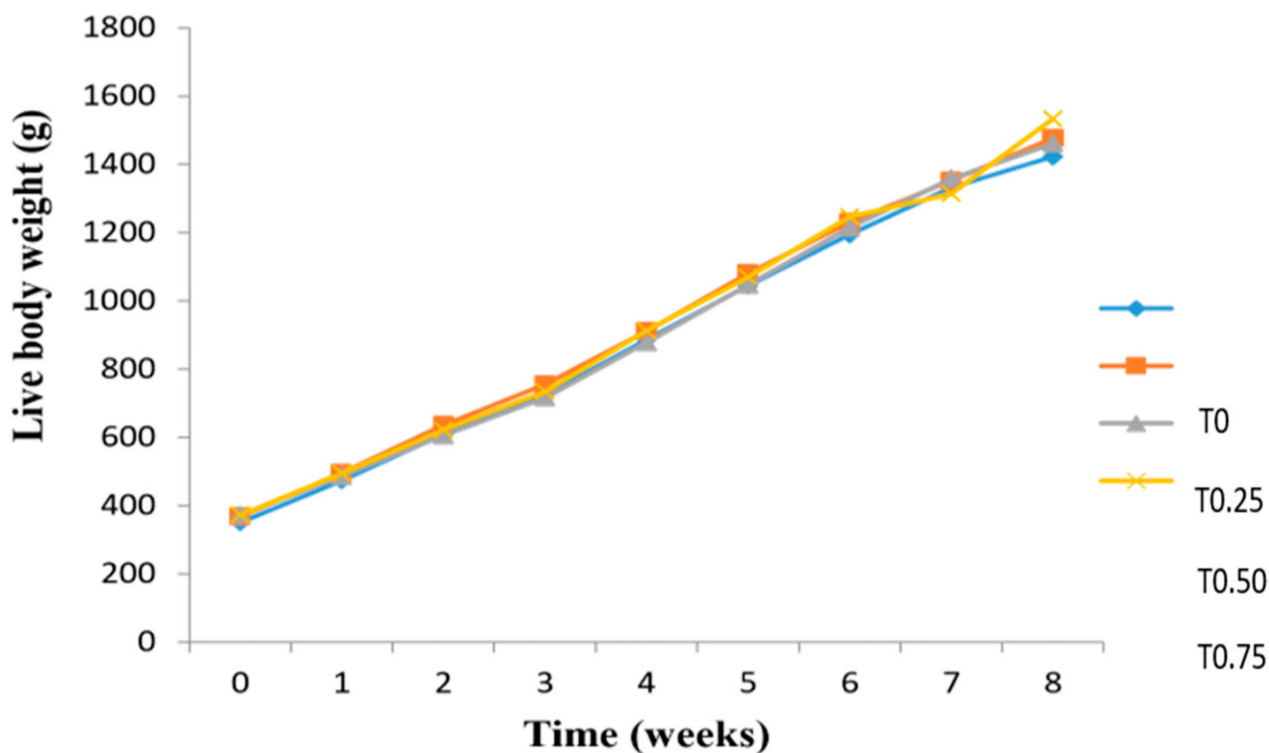


Figure 2. Live body weight evolution as influenced by the level of *D. edulis* leaf powder in feed of Brahma hens. T0: Control (0% additive); T0.25: Control diet containing 0.25%; T0.50: Control diet + 0.5%; T0.75: Control diet + 0.75 of *D. edulis* leaves powder respectively.

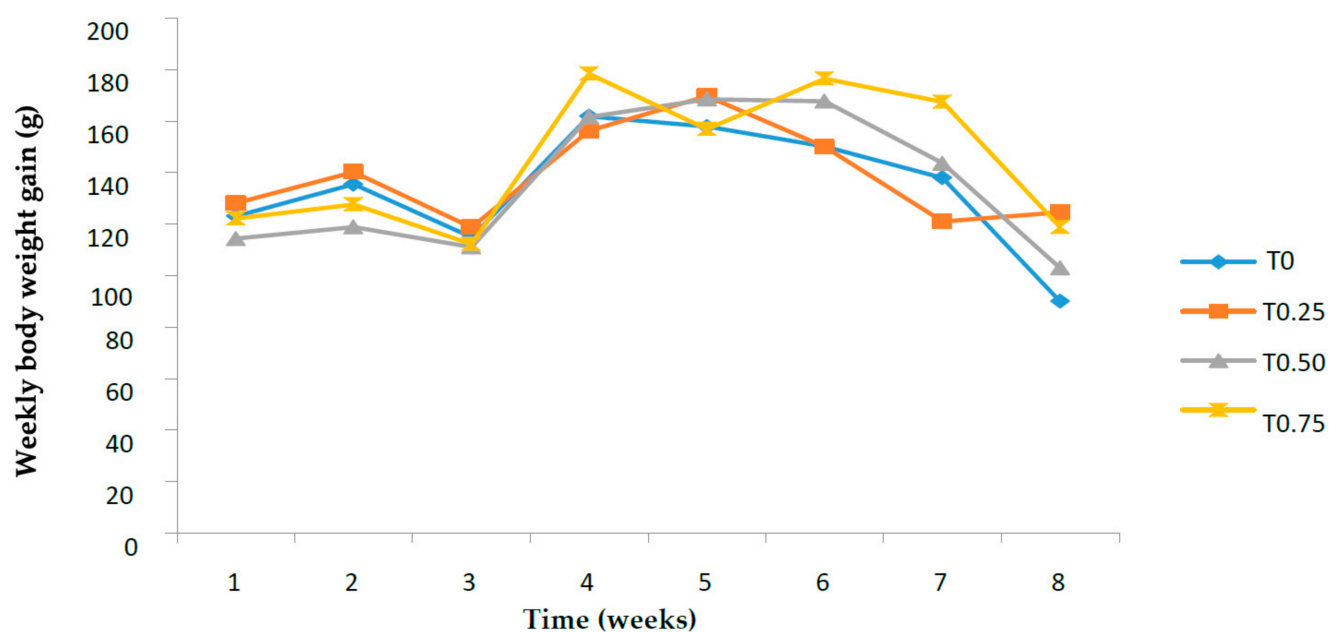


Figure 3. Evolution of weekly body weight gain as influenced by the level of *D. edulis* leaf powder in feed of Brahma hens. T0: Control (0% additive); T0.25: Control diet containing 0.25%; T0.50: Control diet + 0.5%; T0.75: Control diet + 0.75 of *D. edulis* leaves powder respectively.

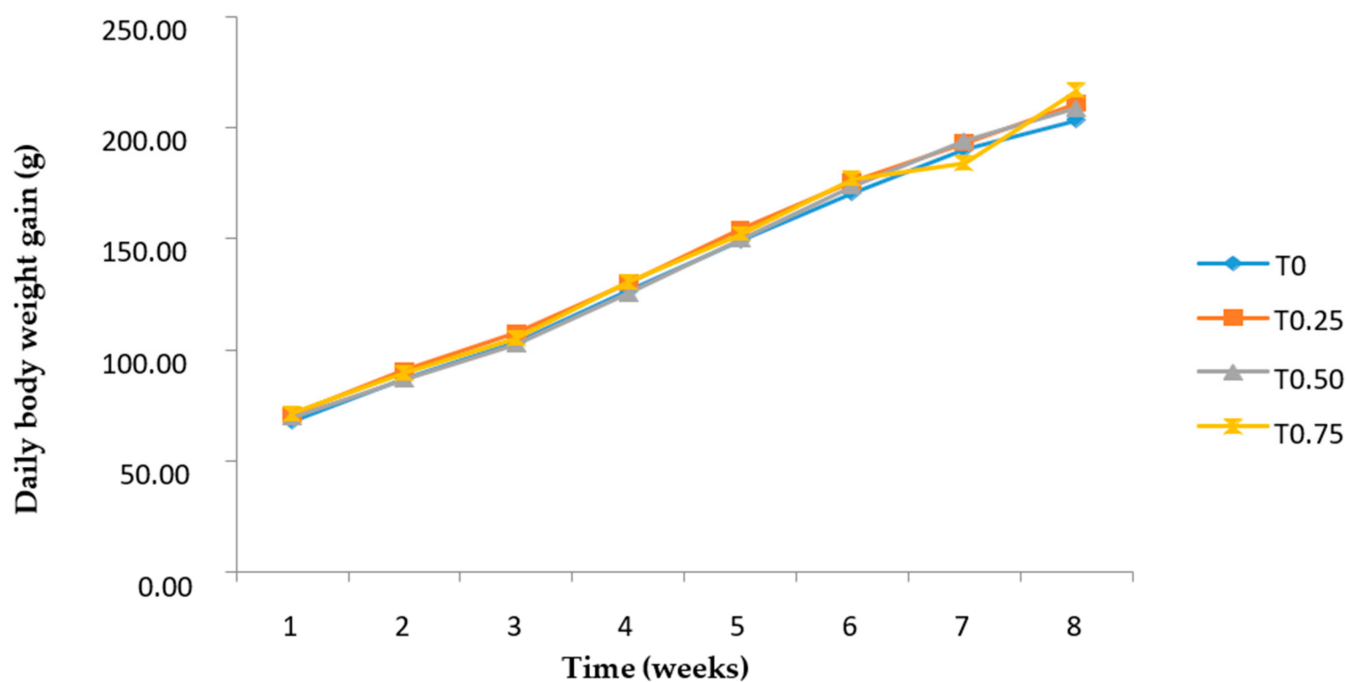


Figure 4. Evolution of daily body weight gain as influenced by the level of *D. edulis* leaf powder in feed of Brahma hens. T0: Control (0% additive); T0.25: Control diet containing 0.25%; T0.50: Control diet + 0.5%; T0.75: Control diet + 0.75 of *D. edulis* leaves powder, respectively.

Effects of *D. edulis* Leaf Powder on Feed Consumption in Brahma Hens

Figure 1 illustrates the effect of *Dacryodes edulis* leaf powder in Brahma hens on feed consumption. The curve shows the same profile and tendency and, moreover, the lowest feed consumption was observed with 0.25% *D. edulis* leaves powder in feed.

Effects of *D. edulis* Leaf Powder in Feed on Live Body Weight in Brahma Hens

The evolution of live body weight according to the rate of *D. edulis* powder leaves in Brahma hens is illustrated in Figure 2. It appears that there was a general increase in live body weight from the start until the end of the trial, whatever the treatment.

Effects of *D. edulis* Leaf Powder in Feed on Body Weight in Brahma Hens

The body weight evolution according to the rate of *D. edulis* powder leaves in Brahma hens is shown in Figure 3. Body weight evolution showed the same profile and tendency in all the treatment groups from the start until the end of the trial. Nevertheless, an increasing tendency was observed with increasing doses of *D. edulis* powder leaves.

Effects of *D. edulis* Leaf Powder in Feed on Daily Body Weight Gain in Brahma Hens

Figure 4 shows the evolution of daily weight gain according to *D. edulis* leaf powder in feed of brahma hens. The curve shows the same profile and tendency in daily weight gain evolution throughout the trial period. It appears that, from the beginning of the test period until the 7th week, the daily weight gain increased regularly, regardless of the treatment.

Effects of *D. edulis* Leaf Powder in Feed-on-Feed Conversion Ratio (FCR) in Brahma Hens

The evolution of feed conversion ratio in birds fed with *D. edulis* powder leaves in feed of Brahma hens is illustrated on Figure 5. The curve shows an irregular evolution of feed consumption in all the treatment groups. Moreover, the lowest FCR at the 8th week was observed with 0.75% *D. edulis* leaves powder, and the highest was observed with 0% in feed.

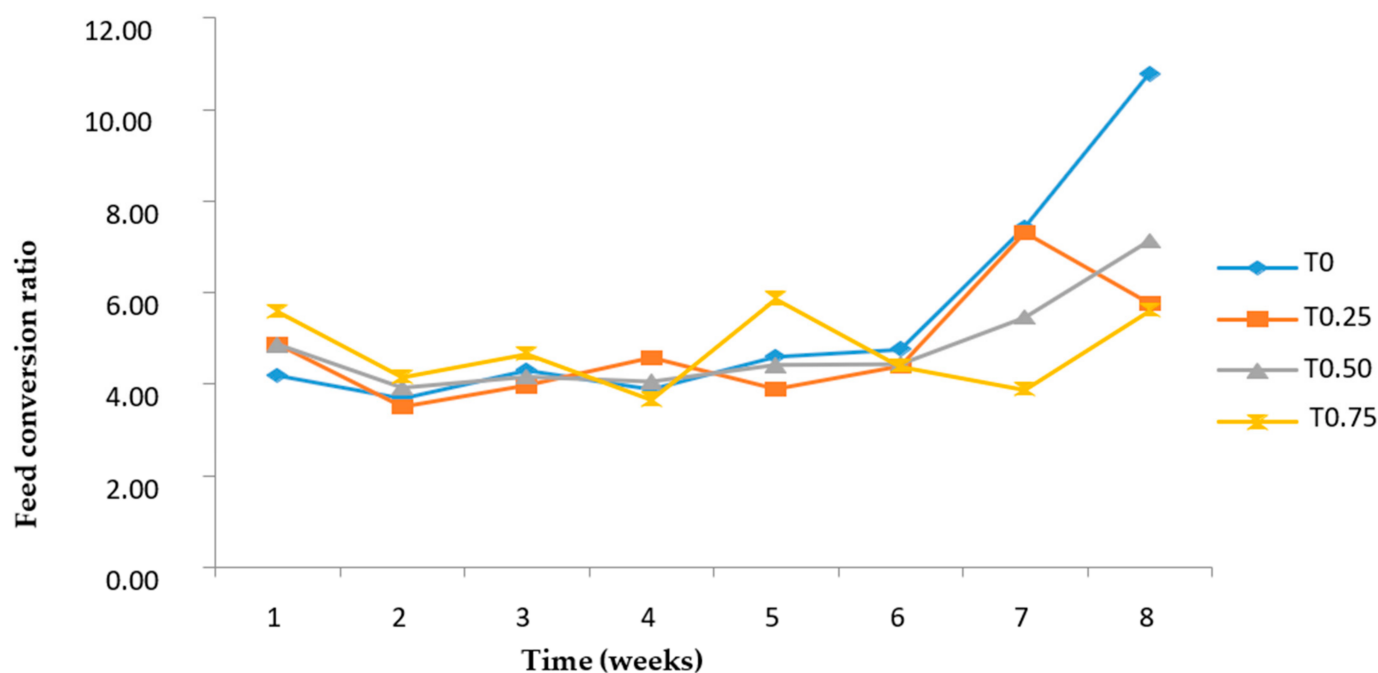


Figure 5. Evolution of feed conversion ratio as influenced by the level of *D. edulis* leaf powder in feed of Brahma hens. T0: Control (0% additive); T0.25: Control diet containing 0.25%; T0.50: Control diet + 0.5%; T0.75: Control diet + 0.75 of *D. edulis* leaves powder, respectively.

Effects of *D. edulis* Leaf Powder in Feed-on-Feed Efficiency in Brahma Hens

Figure 6 shows the evolution of feed efficiency (FE) on Brahma birds fed with *D. edulis* leaf powder at different concentrations. It shows that the FE decreased regularly, whatever the concentration of *D. edulis* leaf powder was in the feed.

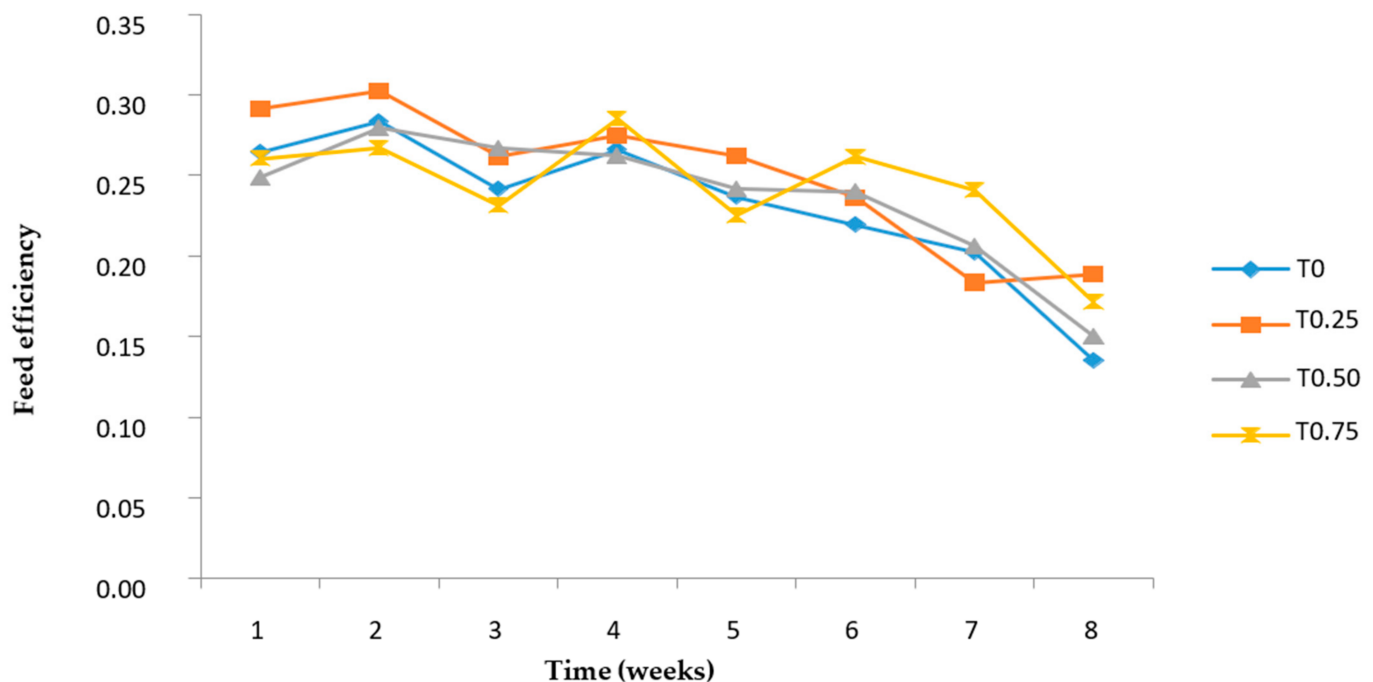


Figure 6. Evolution of feed efficiency as influenced by the level of *D. edulis* leaf powder in feed of Brahma hens' feed. T0: Control (0% additive); T0.25: Control diet containing 0.25%; T0.50: Control diet + 0.5%; T0.75: Control diet + 0.75 of *D. edulis* leaves powder, respectively.

Table 3 summarizes the feed intake, live body weight, weekly body weight gain, daily weight gain, feed conversion ratio, and feed efficiency at the end of the sixty days trial.

As shown on the table, *D. edulis* leaves powder induced a significant ($p < 0.05$) increase in feed consumption at all the tested percentages compared to the control group. Birds exposed to 0.5% and 0.75% *Dacryodes edulis* leaf powder in their feed had a higher feed consumption than birds exposed to 0.25% *Dacryodes edulis* leaves powder in their feed.

The effects of *D. edulis* powder leaves on live body weight are shown on Table 3. There was an increase in live body weight of Brahma birds exposed to *D. edulis* leaves powder compared to the control. Meanwhile, this increase was significant ($p < 0.05$) only in birds fed with *D. edulis* leaf powder at 0.75% compared to the control group.

As reported in Table 3, Brahma birds fed with *D. edulis* powder leaves, regardless of the concentration, had no significant ($p > 0.05$) effect on body weight when comparing exposed birds to those of control. Nevertheless, body weight increased in *D. edulis* leaf powder treated birds.

As shown on Table 3, at 60th day of experiment, DWG was not significantly affected, whatever the concentration this of *D. edulis* powder leaves in Brahma hen feed.

At the end of the test, the average FE recorded between the different levels of *D. edulis* leaf powder in feed were comparable ($p > 0.05$), although it was slightly higher in birds having received the ration with 0.75% *D. edulis* leaf powder and lower in birds that received 0.25% *D. edulis* leaf powder in the feed (Table 3).

Figure 7 shows the relationships between feed intake and graded concentration of *D. edulis*. It resulted in the variation of feed intake due the incorporation of *D. edulis* powder in broiler feed is at 66.39% following the polynomial regression.

Figure 8 shows the relationships between live body weight and graded concentration of *D. edulis*. It resulted in a high relation between live body weight and *D. edulis* concentration. As shown in the shape, 62.98% of variation observed in live body weight is related to *D. edulis* power in feed.

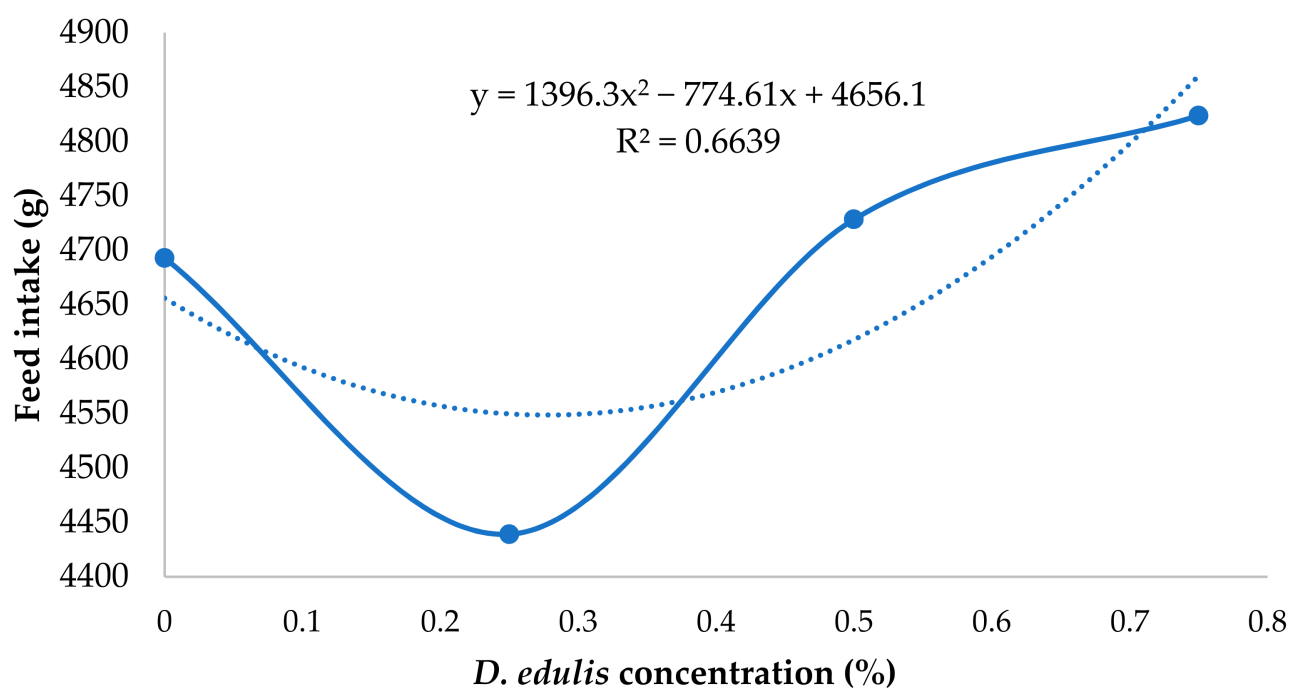


Figure 7. Regression curve of feed intake and graded concentration of *D. edulis*.

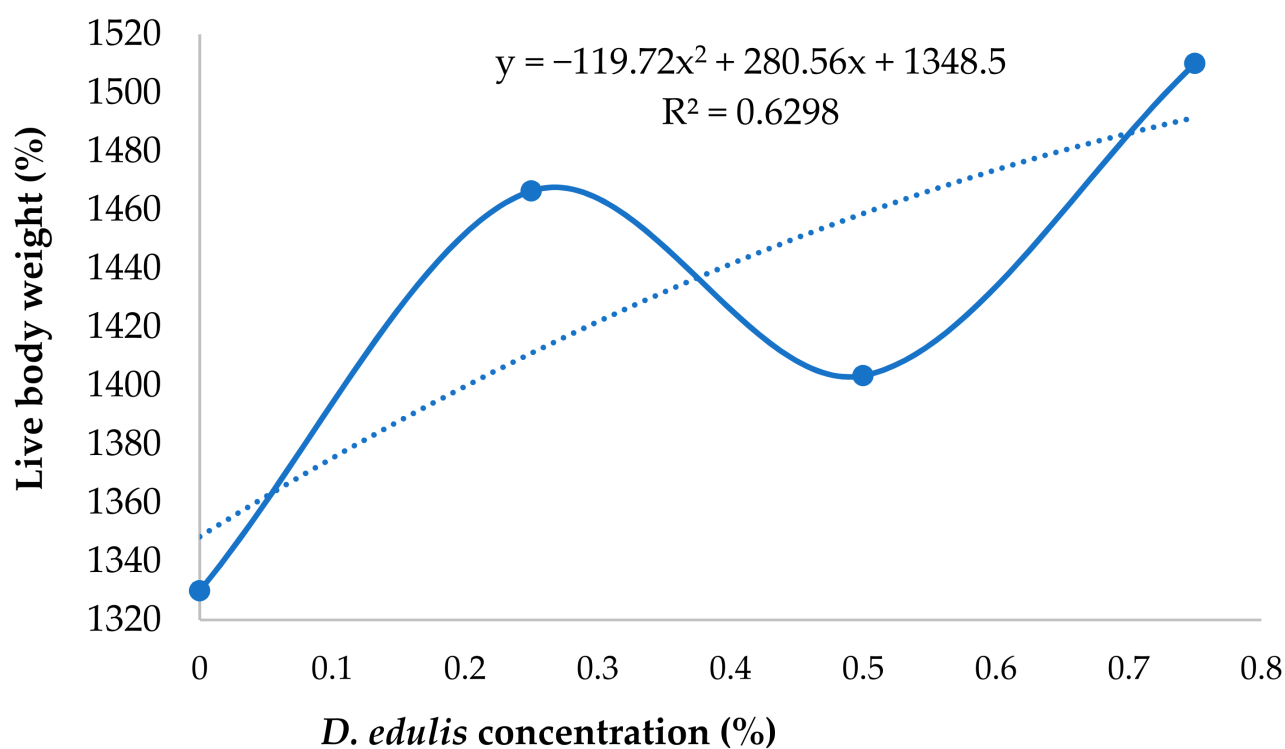


Figure 8. Regression curve of live body weight and graded concentration of *D. edulis*.

Figure 9 illustrate the relationships between live body weight and graded concentration of *D. edulis*. From the figure, it was observed that feed conversion ratio decreases at 78.48% with the inclusion of *D. edulis* powder, whatever the concentration in broiler feed.

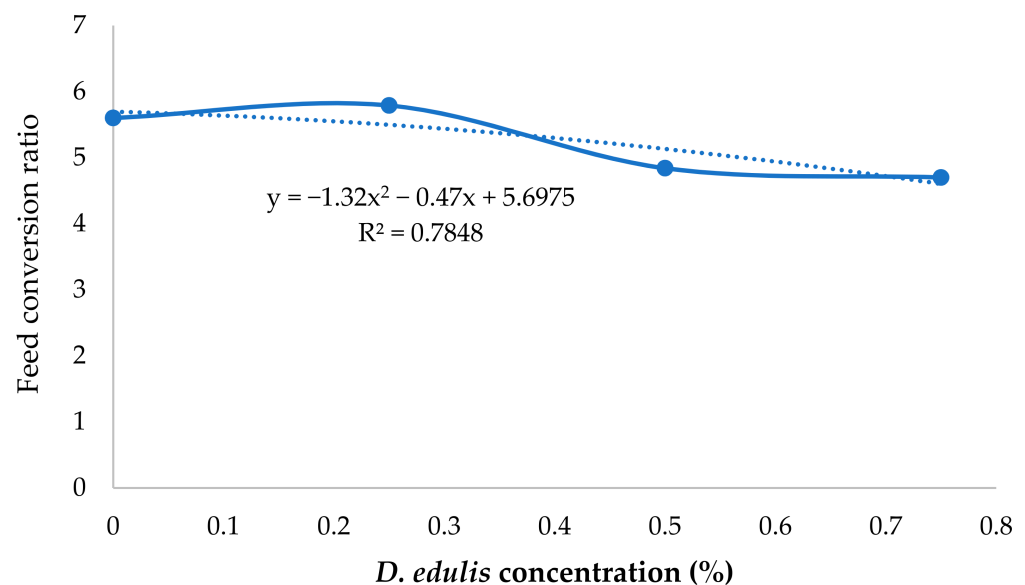


Figure 9. Regression curve of feed conversion ratio and graded concentration of *D. edulis*.

3.1.2. Effects of *D. edulis* Leaves Powder in Feed on Organs Characteristics of Brahma Hens

Table 4 shows the effects of *D. edulis* powder leaves in feed on organs characteristics of Brahma hens. *D. edulis* powder leaves did not significantly ($p > 0.05$) affect relative weight of the liver, heart, volume of the liver and intestinal density when comparing exposed birds to the control. However, slight decreases were noted in birds exposed to *D. edulis* leaves powder in all tested concentration for relative weight of the liver and the heart.

Table 4. Effects of *D. edulis* leaves powder in feed on organs characteristics of Brahma hens.

Organs	T0	<i>D. edulis</i> Concentration (%)			<i>p</i> Value
		T0.25	T0.50	T0.75	
R W Liver (%)	2.01 ± 0.17	1.92 ± 0.48	1.90 ± 0.39	1.73 ± 0.26	0.61
R W Heart (%)	0.49 ± 0.05	0.41 ± 0.05	0.46 ± 0.09	0.48 ± 0.11	0.26
R W abdominal Fat (%)	3.32 ± 0.81 ^a	3.00 ± 0.67 ^{ab}	2.21 ± 0.72 ^{bc}	1.68 ± 0.46 ^c	0.01
Liver Volume (mL)	24.28 ± 2.83	24.00 ± 5.34	23.00 ± 3.75	24.40 ± 3.36	0.92
Intestinal density (g/cm)	0.35 ± 0.052	0.31 ± 0.04	0.34 ± 0.08	0.37 ± 0.10	0.55

^{a,b,c}: values affected with the same letter in the same line are not significantly different ($p > 0.05$); R W: Relative weight; T0: 0% additive; T0.25: Control diet containing 0.25%; T0.50: Control diet + 0.5%; T0.75: Control diet + 0.75 of *D. edulis* leaves powder respectively; *P*: probability.

The relative weight of abdominal fat in birds fed with feed recorded a dose-dependent decrease, though this decrement was only significant ($p < 0.05$) at 0.5% and 0.75% of *D. edulis* with respect to those given non supplemented feed.

3.1.3. Effects of *D. edulis* Leaves Powder in Feed on Parameters of Oxidative Stress of Brahma Hens

Table 5 summarizes the effects of *D. edulis* leaves powder in feed on parameters of oxidative stress in Brahma hens. *D. edulis* leaf powder did not significantly ($p > 0.05$) affect serum activities of SOD, CAT, total peroxidase, and MDA level of exposed birds compared to those of control. However, a slight decrease in serum SOD, CAT and MDA was noted in *D. edulis* treated birds in all the tested concentrations compared to the control.

Table 5. Effects of *D. edulis* leaves powder in feed on parameters of oxidative stress of Brahma hens.

Organs	Control T0	<i>D. edulis</i> Concentration (%)			<i>p</i> Value
		T0.25	T0.50	T0.75	
SOD (U/min/g TP)	3.96 ± 0.75	3.67 ± 0.41	3.55 ± 0.49	3.24 ± 0.35	0.28
CAT (μM/min/g TP)	0.85 ± 0.18	0.78 ± 0.14	0.69 ± 0.85	0.75 ± 0.12	0.26
T Peroxidase (mM/min/g TP)	0.009 ± 0.001	0.010 ± 0.001	0.011 ± 0.004	0.008 ± 0.001	0.28
MDA (μM/mL)	0.62 ± 0.06	0.60 ± 0.04	0.59 ± 0.07	0.56 ± 0.06	0.21

SOD: Superoxide dismutase; CAT: Catalase; MDA: Malondialdehyde; TP: Total protein; T peroxidase: Total peroxidase; T0: Control (0% additive); T0.25: Control diet containing 0.25%; T0.50: Control diet + 0.5%; T0.75: Control diet + 0.75 of *D. edulis* leaves powder respectively; P: probability.

3.1.4. Effects of *D. edulis* Leaves Powder in Feed on Serum Biochemical Concentrations of Brahma Hens

Table 6 presents the variation of serum biochemical concentrations depending on the level of *D. edulis* powder leaves in feed of Brahma hens. It was observed that *D. edulis* powder leaves in all tested concentration did not significantly ($p > 0.05$) affect ALT activity, creatinine, and urea serum content. On the other hand, *D. edulis* powder leaves in feed of Brahma birds in all tested concentration led to a decrease in activities of Aspartate aminotransferase (AST) in serum. Moreover, a significant ($p < 0.05$) decrease in serum AST concentration in exposed birds was observed only at 0.75% concentration compared to the control group.

Table 6. Serum biochemical concentrations depending on the concentration of *D. edulis* powder leaves.

Biochemical Parameters	Control T0	<i>D. edulis</i> Concentration (%)			<i>p</i> -Value
		T0.25	T0.50	T0.75	
ALT (U/L)	51.04 ± 12.89	41.56 ± 13.61	49.22 ± 13.32	43.75 ± 16.54	0.65
AST (U/L)	185.75 ± 36.99 ^a	150.28 ± 30.50 ^{ab}	143.12 ± 38.65 ^{ab}	119.75 ± 37.38 ^b	0.04
Creatinine (mg/dL)	0.05 ± 0.04	0.03 ± 0.01	0.03 ± 0.01	0.02 ± 0.01	0.28
Urea (mg/dL)	26.63 ± 0.77	26.30 ± 0.63	26.26 ± 0.48	25.93 ± 1.05	0.47

^{a,b}: values affected with the same letter in the same line are not significantly different ($p > 0.05$). ALT: Alanine aminotransferase; AST: Aspartate aminotransferase; T0: Control (0% additive); T0.25: Control diet containing 0.25%; T0.50: Control diet + 0.5%; T0.75: Control diet + 0.75 of *D. edulis* leaves powder, respectively.

3.1.5. Effect of *D. edulis* Leaves Powder in Feed on Histology of the Kidney

Figure 10 illustrates the effect of *D. edulis* powder leaves in feed on histology of the kidney. Histological studies supported the biochemical findings. It can be seen that the histology of the kidney of birds of the control group showed a slight alteration of nephrocytes. Furthermore, incorporation of *D. edulis* powder leaves in the diet, whatever the concentration, showed amelioration of these alterations, with this amelioration being more pronounced with 0.25% *D. edulis* leaves powder in the diet, and it equally shows a distinct and well organized arrangement of the glomerulus with its bowman capsule, indicating the nephroprotective effect of *D. edulis* powder leaves.

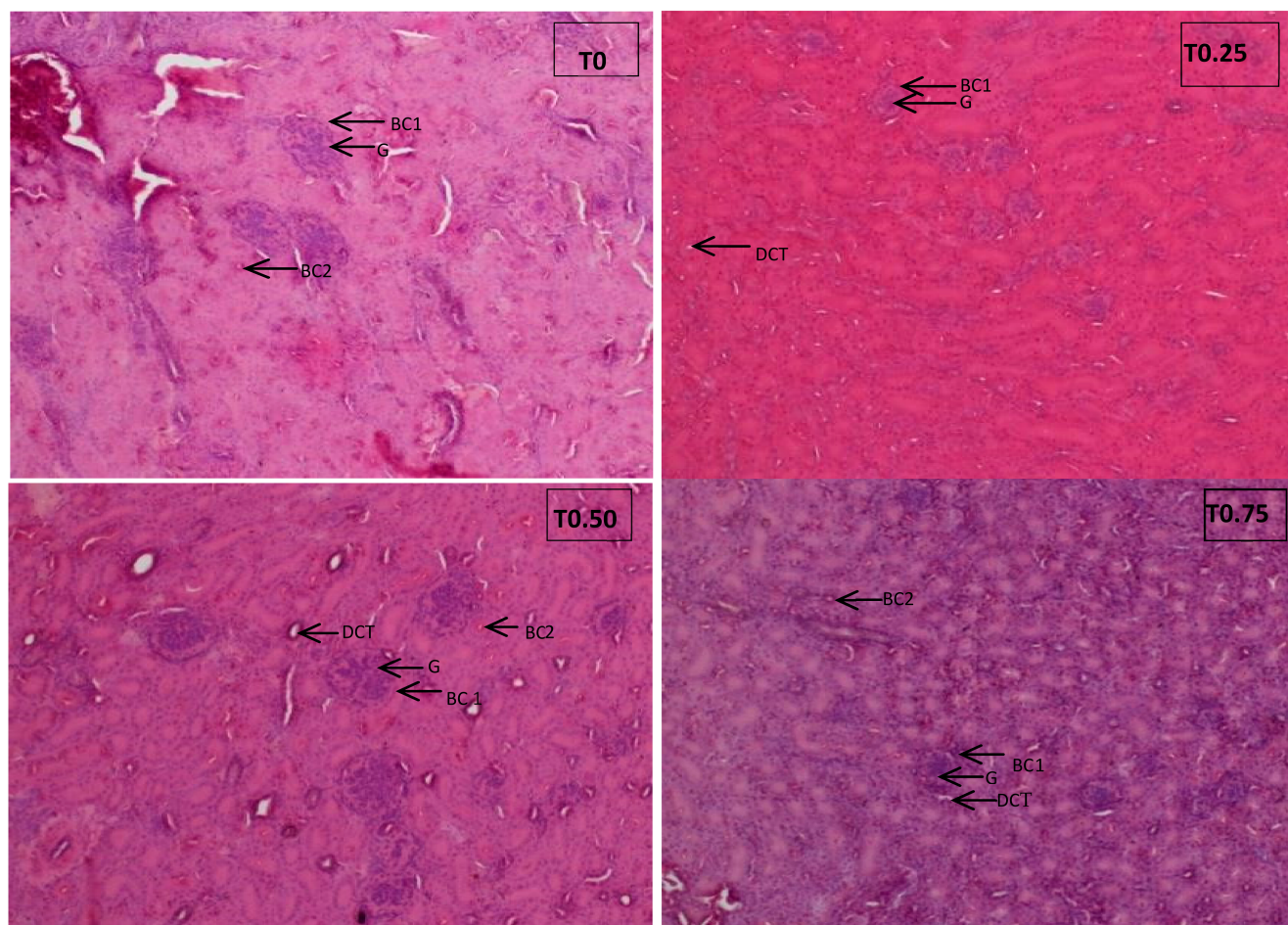


Figure 10. Effects of *D. edulis* on histological structure of the kidney. G: Glomerulus, BC1: Bowman capsule, BC2: Blood cell, DCT: Distal convoluted tubule. T0: Control (0% additive); T1: Control diet containing 0.25%; T2: Control diet + 0.5%; T3: Control diet + 0.75 of *D. edulis* leaves powder, respectively.

3.1.6. Relationship between Growth Parameters, Oxidative Stress, and Toxicity

Some correlations between growth characteristics, oxidative stress, and toxicity are illustrated on Table 7. It was noted that:

Table 7. Correlations between some evaluated characteristics in Brahma hens.

Parameters	FI	LBW	DWG	RW Liver	AST	Creatinine	Urea
LBW	0.95	1					
DWG	0.95 *	0.31	1				
RW Liver	−0.49	−0.87	−0.73	1			
AST	−0.32	−0.90	−0.56	0.95	1		
Creatinine	−0.18	−0.94	−0.44	0.91	0.99 *	1	
Urea	−0.37	−0.92	−0.62	0.98 *	0.99 **	−	1
MDA	−0.51	−0.85	−0.73	0.99 **	0.97 *	0.93	0.99 *

ALT: Alanine aminotransferase; AST: Aspartate aminotransferase; MDA: Malondialdehyde, R W: Relative weight; Daily weight gain; LBW: Live body weight; FI: Feed intake; * Correlation is significant at the 0.01 level (2-tailed); ** Correlation is significant at the 0.05 level (2-tailed)

Negative and non-significant correlations were recorded between the relative weight of the liver and live body weight ($R = -0.87$; $p > 0.05$), the level of malondialdehyde and daily weight gain ($R = -0.73$; $p > 0.05$), malondialdehyde and feed intake ($R = -0.51$; $p > 0.05$), and malondialdehyde and live body weight ($R = -0.85$; $p > 0.05$).

However, daily weight gain was positively and significantly correlated to feed intake ($R = +0.95^*$; $p < 0.01$). Relative weight of the liver was positively and non-significantly correlated with the serum activity of aspartate aminotransferase (AST) ($R = +0.95$; $p > 0.05$).

The level of MDA was positively and significantly correlated to the relative weight of the liver ($R = +0.99^{**}$; $p < 0.05$) and to the activity of aspartate aminotransferase (AST) ($R = +0.97^*$; $p < 0.01$).

3.2. Discussion

The results obtained in the present study reveal that *D. edulis* leaf powder supplementation at 0.5 and 0.75% in feed has resulted to an increase in feed intake in Brahma birds compared to the control group. These results are in contradiction with those obtained by Tangomo [11], who revealed that 0.5 and 1.0% of *D. edulis* leaves powder in feed did not significantly ($p < 0.05$) affect feed intake in local chickens. These results are in line with those obtained by Martínez et al. [26], who reported that addition of *Anacardium Occidentale* L. Leaves Powder at 0.5 and 0.75% in feed induces a significant increase in feed intake in chicken during 10 days of age. This increase in feed consumption observed in the present study could be explained by the fact that phytoadditives improve the flavor, texture, and palatability of feed, thus increasing voluntary feed intake and subsequently the weight gain [27].

Live body weight during the experimental study was significantly ($p < 0.05$) higher with 0.75% *D. edulis* leaves powder diets as compared to control group. This result is in line with that of Tangomo [11], who revealed that 0.5 and 1.0% of *D. edulis* leaves powder in broiler chickens feed increases body weight and weight gain relative to control. The increase in live body weight, body weight, and daily weight gain could be justified by the presence in *D. edulis* leaves powder of bioactive molecules (total phenol, flavonoids) with antimicrobial and antioxidant properties. Phytochemical compounds that have antimicrobial property inhibit the proliferation of intestinal bacteria, such as *Clostridium perferingens*, *E. coli*, *Enterobacter cloaceae*, and *Bacteriodes fragilis* [9], and they increase the bioavailability of nutrients used for growth. Antimicrobial activity would help to limit inflammatory reactions costly in energy in favor of growth [28,29]. On the other hand, molecules with antioxidant properties would protect cells structure against detrimental effects of reactive oxygen species. This effect would increase the cell thickness linking to high body weight. In 0.75% *D. edulis* leaves, powder-fed Brahma birds, standard deviation were very low as compared to those of the rest of the treatments, especially the control group. This result would be linked to the homogeneity in body weight of the birds in the group.

From the results obtained, a non-significant effect was observed in weight of the liver and heart. Inversely, a significant decrease was noted in weight of the abdominal fat in birds exposed to 0.5 and 0.75% of *D. edulis* leaf powder with respect to control. This result is in accordance with those observed by Emadi and Kermanshashi [30], who observed a significant ($p < 0.05$) decrease in abdominal fat with turmeric rhizome powder (0.75% and 0.5%) incorporated in broiler diets. This result is equally in line with the investigation of Tangomo [11] in broiler chickens fed on rations with 0.5 and 1.0% of *D. edulis* leaf powder. To the opposite, Martínez et al. [27] observed a non-significant effect of *Anacardium Occidentale* L. leaf powder at the concentration of 0.5 and 0.75% in diet on liver and heart weights of Broiler chickens. The reduction in the percentage of abdominal fat may be attributed to the presence of tannins in the plant, which inhibits fat absorption in the intestine, and they have been reported to possess lipid lowering effects [31].

Blood parameters are considered as principal physiological, pathological, and nutritional indices that permit the appreciation of the state of an organ. The level of AST and ALT in serum informs the state of hepatic cells [32]. Increase in these enzymes in blood translates hepatocellular damage [33], resulting from the body's response to stress. Incorporation of *D. edulis* leaf powder in the feed of Brahma birds induced a non-significant decrease ($p > 0.05$) in serum ALT activity, but significant ($p < 0.05$) decrease in serum AST activity of *D. edulis*-treated birds. This result is in agreement with those obtained by Tugiyanti

et al. [34], who reported that supplementing avocado seed powder to culled female quails had no significant ($p > 0.05$) decrease on ALT, but a significant ($p < 0.01$) decrease in AST level in reference to the control. A decrease in Serum transaminases (ALT and AST) concentration, although non-significant, in ALT could be linked to the hepatoprotective activity of *D. edulis* leaf powder, implying that *D. edulis* leaf powder was not toxic to the liver. This effect would equally indicate normal functioning of the hepatic tissues. In fact, incorporation of *D. edulis* powder leaves in the diet, whatever the concentration, showed distinct and well organized arrangement of the glomerulus with its Bowman capsule. The hepatoprotective properties of *D. edulis* observed in the study may be related to its high content of antioxidant compounds, such as flavonoids and phenols.

4. Conclusions

At the end of this study, which aimed at evaluating the growth characteristics, oxidative stress, and toxicity in *Gallus gallus* domesticus (Brahma) hens fed with a diet containing *Dacryodes edulis* (Safou) leaves powder, the following conclusion were drawn: incorporation of *D. edulis* leaf powder in the diet of Brahma hens resulted in an increase in feed intake and live body weight of Brahma hens; it led to the prevention of oxidative stress, indicated by a non-significant effect on MDA. It also showed a non-toxicity effect on the liver and kidneys, since there was a decrease in activities of transaminase, as well as a non-significant impact on creatinine and urea levels in the serum with *D. edulis* leaf powder in diets. More research on *D. edulis* can be conducted on broiler feed at 0.75% concentration, which will show a significant increase and decrease, respectively, in live body weight and serum aspartate aminotransferase activity.

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