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GROWTH PERFORMANCE, CARCASS CHARACTERISTICS, IMMUNE RESPONSE AND MEAT QUALITY OF BROILER CHICKENS FED *MUNTINGIA CALABURA* LEAF EXTRACT

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Abstract. This experiment was carried out to determine the effect of feeding *Muntingia calabura* leaf extract on the growth performance, carcass characteristics, immune response and meat quality of broiler chickens. 500 1-day old broiler chicks (Cobb 500) were randomly distributed into five treatments of five replicates consisting of ten birds each in a completely randomized design model. Birds in treatment one (control) was fed basal diet alone which is adequate in all nutrients according to the recommendation of broilers while treatment two, three, four and five were fed same diet with *Muntingia calabura* leaf extract at 0.3 mL, 0.6 mL, 0.9 mL and 1.2 mL per liter of water correspondingly. The duration of the experiment was 56 days and birds had unrestricted access to feed and fresh water. Body weight gain was higher in birds fed treatment four (2555.3 g) and five (2646.1 g), intermediate in treatment two (2346.6 g) and three (2365.1 g) and lower in treatment one (1948.8 g) ($P<0.05$). Total feed intake and dressing percentage was higher among birds fed *Muntingia calabura* leaf extract relative to the control ($P<0.05$). Conversely, mortality of 2.5 % was recorded in control followed by treatment two with 1.00 % none was recorded in the other treatment ($P<0.05$). Results on the breast muscle revealed that saturated and unsaturated fatty acid was 29.80 %, 41.83 % in control, treatment two (28.11 %, 55.94 %), three (27.19 %, 56.97 %), four (23.15 %, 73.69 %) and five (22.90 %, 73.74 %) in each case ($P<0.05$). Immunoglobulin A, G and M values were influenced ($P<0.05$) by the treatments. In conclusion, feeding *Muntingia calabura* leaf extract up to 1.2 mL/liter of water had no negative effect on the performance and health status of birds.

Keywords: *Muntingia calabura leaf extract, growth, immune response, phytochemicals, medicinal plants, food safety.*

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1. Introduction

The topic of antibiotic growth promoters and their replacement continues to be of great interest to poultry producers with a wide range of options now available to those who are either prevented from using antibiotic growth promoters by local regulations or prefer to use an alternative due to their non-toxic effect (Liz, 2020; John, 2024). The use of medicinal plants is among the key alternative solutions to antibiotics because they possess phytochemicals that has high therapeutic properties and no withdrawal period (John, 2024; Ojediran, 2024).

Muntingia calabura popularly known as 'Jamaican cherry' is a rapidly growing small, evergreen plant belonging to family muntingiacae and order – Malvales. The

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plant is native to southern Mexico central America and western south America. It is also widely cultivated in Asia including India (Sani *et al*., 2012). The leaves are soft evergreen and alternate, ablong phyllotaxy about 4-15 cm long and 1-6 cm wide, oblong with toothed margin oblique at the base and characterized with dark green color and upper surface indicates presence of minute hair (Su *et al*., 2003).

Reports on the pharmacological evaluation of *Muntingia calabura* leaf has shown that it possesses numerous phyto-constituents such as: flavonoids, terpenoids, tannins, alkaloids, saponins, amongst others (Chen *et al*., 2007) which performs multiple biological activities: antimicrobial, antioxidant, anti-helminthic, antifungal, antiviral, hepato-protective, antibacterial, anti-tumor, cytotoxic, anti-diuretic and hypolipidemic (Chen *et al*., 2004). *Muntingia calabura* is popular in folklore medicine and has been used for the treatment of severe diarrhea, dysentery, urethral discharge, gastro-intestinal infection, headache, dermatitis and other bacterial infections (Balan *et al*., 2013; Zakaria *et al*., 2006a)

The aqueous extract of leaf has been reported to contain volatile compounds such as myrcene, thymol, α terpinol, linalool, geraniol, nerol, citronellol, eugenol, α lonone, β sitosterol, α Amyrin, Lupelol, α tocopherol and β carotene (Preethi *et al*., 2010; Zakaria *et al*., 2006b). In a study performed by Ibrahim et al. (2012), the ethanolic extract of the different parts of *Muntingia calabura* demonstrated antimicrobial activity against *Escherichia coli, Pseudomonas aeruinosa, Candida albicans, Streptoccoccus spp, Salmonella spp* and *Aspergillus spp*. It also showed that the extracts from their flowers demonstrated more antiseptic and antiplasmodic properties than the other parts of the plant.

Previous findings by notable researchers have shown that dietary supplementation of medicinal plants in broilers can positively influence their growth, gastro intestinal tract morphology and physiology immune response and meat quality (Alipour *et al*., 2015; Oloruntola *et al*., 2018; Alagbe *et al*., 2024); Mgbemena *et al*., 2019); Gou *et al*., 2004). However, there is little or no report on the dietary supplementation of *Muntingia calabura* leaf extract in broilers. This information is timely, because livestock producers have turned against the use of antibiotics due to the increasing cases of antimicrobial resistance and their health effect. This studies will further promote organic agriculture and food safety.

Therefore, this study was carried out to ascertain the effect of feeding *Muntingia calabura* leaf extract on the growth performance, carcass characteristics, immune response and meat quality of broilers.

2. Materials and methods

Experimental station, ethical approval and extraction of Muntingia calabura

The trial was conducted at Sumitra Institute's poultry unit situated between 26^{o} 17' N and 70^{o} 47' E. Experiment was executed in accordance to the specifications laid down by ethics council of Sumitra Research Institute, Gujarat India in the month of September to November, 2023 (CN/117H/2023).

Fresh leaves of Muntingia calabura were collected within Sumitra Institute, Gujarat in the month of September, 2023 and sent to the Crop Science section for proper identification by a certified taxonomist (Dr. Singh Liu) for proper authentication. It was later assigned a reference number (JP/008/223), shade dried for six days with an electronic blender. Three hundred grams of grounded Muntingia calabura was soaked in

1 liter and boiled at seventy ℃ for ten minutes, kept for three days before it was sieved using a Whatman filter paper to obtain Muntingia calabura leaf extract and kept in the refrigerator at four ℃. 200 milliliters of Muntingia calabura leaf extract was sent to the department of biochemistry, Sumitra Research Institute, Gujarat for further evaluation.

Animal management, experimental diet and design

500 1-day old broiler chicks (Cobb 500) were purchased from a commercial farm in Gujarat in the month of September, 2023 and moved to the poultry unit of Sumitra Research Institute, India. On arrival, birds were unboxed and the average initial weight was recorded using a digital scale before it was randomly distributed into five treatments of five replicates consisting of ten birds each. Birds were raised in an environmentally controlled battery cage equipped with automatic nipple drinker's, feeders measuring 200 cm by 150 cm by 60 cm (length × width × height) in three tiers per row and given anti-stress (Glucomol wsp® at 10 grams/10 liter). Continuous lighting system was provided to enable birds to have unrestricted access to feed and water. Basal diet was sufficient in all nutrients according to the recommendation of National Research Council in 1994. Birds in treatment one was fed basal diet without Muntingia calabura leaf extract while treatment two, three, four and five were fed same diet with Muntingia calabura leaf extract at 10 milliliters, 20 milliliters, 30 milliliters and 40 milliliters per liter of water correspondingly. A completely randomized design was adopted throughout the trial which lasted for sixty days.

Collected data

Body weight gain was calculated as the difference between the final body weight and initial body weight. Feed consumption was determined as the difference between the feed offered and refused. Feed conversion ratio (feed consumed to produce a unit of gain) was computed as the ratio of average daily feed intake to average daily weight gain. Average daily feed intake was estimated by dividing the total feed intake by the duration of experiment in days while average daily weight gain was computed by dividing the weight gain by the duration of experiment in days, all the variables were expressed in grams.

Estimation of phyto-constituents in Muntingia calabura leaf extract

The laboratory procedures for the determination of tannins, flavonoids, terpenoids, alkaloids, saponins and phenolic compounds were carried out according to methods outlined by Alagbe (2024).

Proximate analysis of starter and finisher diet

Experimental diets were analyzed using Foss NIRS^{TM} instrument which uses optical near infra-red technology to ensure precision in result. One hundred and fifty grams of feed sample was placed in the collection pan and the machine was adjusted at a wavelength precision of 400 – 2500 nm, optical band width (9.00 ± 0.2 nm), spectral resolution (0.5 nm) to give a faster result at an analysis time less than one minutes.

Carcass evaluation and measurement

At the end of the trial, ten birds were randomly selected from each treatment for carcass evaluation. Birds were feed starved for 12 hours, placed on clean water and slaughtered manually with a sharp knife and de-feathered by immersing each bird in hot water boiled at 80℃ for fifteen minutes. Carcass were eviscerated removing the internal organs and measured with using a digital scale. Back, wing, thigh, breast and drumsticks were measured with a digital scale as part of the carcass measurement. The dressing percentage was calculated as the proportion of carcass weight to slaughter weight multiplied by hundred.

Fatty acid analysis

Meat from the breast muscle of slaughtered chicken were collected into a sample bottle and placed in an ice pack before it was transferred to the laboratory for analysis. Fatty acid compositions were determined using a commercial kit (Fin analyzer, model – YLB 500 GC, China) following the specified procedures outlined by the manufacturer. 0.5 sodium hydroxide plus 1.5 milliliter methanol was added to each sample and mixed together followed by heating at 100℃ for 5 minutes before it was allowed to cool down at 30 – 40 ℃. Addition of trifluoroborane methanol solution allows the sample to form separate layers before standardization using YLB 500 GC which has the following technical specifications: Injection volume (1µL), oven and capillary temperature (140℃, 250℃) and detector FID (100℃).

Immunoglobulin examination

2 mL of blood samples were collected from ten birds were randomly selected from each treatment at the end of the experiment for immunoglobulin assessment. Test was carried out using a commercial diagnostic kit Immulite ® 1000 system. To ensure precision machine was adjusted according to the manufacturers recommendation, operating temperature was set at 18-32℃ while wavelength band (1000 nm).

Data analysis

Data collected on growth performance, carcass evaluation and fatty acid analysis were analyzed using the general linear model procedures of Statistical Analysis Systems software (SAS). Differences between treatment means were separated using software of the same package. Significant differences were declared at $P<0.05$

Experimental outcome

The presence of tannins (502.58 milligram/gram), flavonoids (820.69 milligram/gram), terpenoids (401.17 milligram/gram), phenolic compounds (1006.5 milligram/gram), alkaloids (115.16 milligram/gram) and saponins (100.8 milligram/gram) were identified in Muntingia calabura leaf extract (Table 2). Phenolic compounds had the greater concentration while alkaloids had the least concentration in order of abundance.

As revealed in Table 2, final body of birds at 0-28 days (starter phase) took the form of 912.5 grams to 1084.9 grams, body weight gain (866.4 – 1084.9 grams per bird), average daily weight gain (30.94 to 38.75 grams per bird), total feed intake (1300.4 – 1300.9 grams), average daily feed intake (46.44 – 46.46 grams per bird), feed conversion ratio (1.20 – 1.50) and mortality (1.00 – 1.50 %). Body weight gain of birds fed diet 2 (0.3 mL *Muntingia calabura* leaf extract) and diet 3 (0.6 mL *Muntingia calabura* leaf extract) were similar ($P>0.05$) to those given diet 4 (0.9 mL *Muntingia calabura* leaf extract) and diet 5 (1.2 mL *Muntingia calabura* leaf extract) but significantly greater than those fed diet 1 (without *Muntingia calabura* leaf extract). *Muntingia calabura* leaf extract had no significant ($P>0.05$) on average daily feed intake while highest ($P<0.05$) mortality was recorded in birds given diet 1 (1.50 %) followed by diet 2 (1.0 %) while none was recorded among birds in diet 3, 4 and 5 correspondingly which also had an excellent feed conversion ratio. At the finisher phase (29 -56 days), average daily weight gain of birds fed diet 4 (0.9 mL *Muntingia calabura* leaf extract) were similar ($P>0.05$) to those in diet 5 (1.2 mL *Muntingia calabura* leaf extract) while those given diet 2 (0.3 mL *Muntingia calabura* leaf extract) and diet 3 (0.6 mL *Muntingia calabura* leaf extract) also followed similar trend ($P>0.05$) but significantly higher than those fed diet 1 (without *Muntingia calabura* leaf extract). Conversely, average daily feed intake were greater among birds fed diet 3, 4 and 5,

intermediate in diet 2 and lowest in diet 1 (P<0.05). In the overall production cycle (0 – 56 days), average daily weight gain values which varied from 34.80 – 47.25 grams per bird were higher among birds fed diet 4 and 5, intermediate in diet 2 and 3, lowest in diet 1 (P<0.05). Average daily feed intake were greater among birds fed *Muntingia calabura* leaf extract relative to those fed diet 1 (P<0.05) which also has the highest mortality of 2.5 % at the end of the production cycle. Feed conversion ratio were significantly (P<0.05) impacted by feeding *Muntingia calabura* leaf extract to birds.

Effect of *Muntingia calabura* leaf extract on the carcass characteristics of broiler chicken (Table 4) revealed that slaughter weight, carcass weight and dressing percentage which took the form of 1971.2 – 2806.3 grams, 1421.5 to 2306.5 grams and 72.11 – 82.19 percent were higher among birds fed diet 4 (0.9 mL *Muntingia calabura* leaf extract) and diet 5 (1.2 mL *Muntingia calabura* leaf extract), intermediate among birds given diet 3 (0.3 mL *Muntingia calabura* leaf extract) and diet 2 (0.3 mL *Muntingia calabura* leaf extract), lowest in diet 1 (without *Muntingia calabura* leaf extract) (P<0.05). Average weights of heads, wing, liver and heart were exactly comparable (P>0.05) in all the groups except for those of legs, breast, thigh, back and gizzard (P<0.05).

Effect of *Muntingia calabura* leaf extract on the fatty acid characterization of meat in the breast muscle of broilers (Table 5) revealed at it contained saturated fatty acid which comprises of myristic acid, palmitic acid, stearic acid, arachidic acid, behenic acid and lignoceric acid values which vary from 2.50 – 3.81 %, 2.31 – 3.00 %, 10.02 – 13.81 %, 5.06 – 5.55 %, 1.82 – 2.71 % and 0.40 – 0.92 % in that order. Palmitoleic acid, oleic acid, erucic acid, gadoleic acid and nervonic acid (monounsaturated fatty acid) took the form of (2.16 – 5.17 %), (2.25 – 3.80 %), (10.64 – 12.71 %), (4.26 – 7.18 %) and (1.55 – 2.87 %) accordingly and polyunsaturated fatty acid; alpha (α)-linolenic acid (10.22 – 20.11 %), gamma (γ) linolenic acid (0.87 – 2.88 %), arachidonic acid (6.11 – 10.56 %), eicosadienoic acid (0.55 – 1.95 %), docosapentaenoic acid (1.97 – 3.84 %) and decosatetraenoic acid (1.25 – 2.67 %). *Muntingia calabura* leaf extract had a significant (P<0.05) on all the variables examined in the experiment.

Effect of Muntingia calabura leaf extract on the Immunoglobulins of broilers presented in Table 6 showed that immunoglobulin A (Ig A), IgG and IgM took the form of 7.66 – 10.26 (µg/mL), 2.29 – 4.15 (µg/mL) and 1.29 – 2.98 (µg/mL) in that order. Ig A values were greater among birds fed diet 4 (0.9 mL *Muntingia calabura* leaf extract) and diet 5 (1.5 mL *Muntingia calabura* leaf extract/litre of water), intermediate in diet 2 (0.3 mL *Muntingia calabura* leaf extract per litre of water) and diet 3 (0.6 mL *Muntingia calabura* leaf extract) and lowest in diet 1 (without *Muntingia calabura* leaf extract) (P<0.05). Conversely, IgG and IgM values were higher among birds fed *Muntingia calabura* leaf extract relative to those without *Muntingia calabura* leaf extract in diet 1 (P<0.05).

Table 1. Ingredients and chemical composition of experimental diet.

	Starter phase (0 – 28 d)	Finisher phase (29 – 56 d)
Ingredients	Quantity (%)	Quantity (%)
Yellow corn	53.00	55.95
Wheat bran	5.00	6.00
Soybean meal	26.50	25.00
Groundnut meal	5.00	4.00
Fish meal (Imported: 72%)	5.00	2.00
Limestone	1.50	2.00
Bone meal	3.00	4.00
Methionine	0.25	0.25
Lysine	0.20	0.25
Mineral/Vitamin Premix	0.25	0.25
Salt	0.25	0.25
Toxin binder	0.05	0.05
Total	100.0	100.0
Analyzed analysis (% dry matter)		
Crude protein (%)	23.47	21.16
Crude fibre (%)	3.56	4.82
Ether extract (%)	3.20	3.83
Calcium (%)	1.49	1.47
Phosphorus (%)	0.58	0.57
Metabolizable energy (MJ/kg)	11.89	12.30

Table 2. Phyto-constituents in Muntingia calabura leaf extract

Chemical components	Units	Concentration
Tannins	Milligram/gram	502.58
Flavonoids	Milligram/gram	820.69
Terpenoids	Milligram/gram	401.17
Phenols	Milligram/gram	1006.5
Alkaloids	Milligram/gram	115.16
Saponins	Milligram/gram	200.8

Table 3. Effect of *Muntingia calabura* leaf extract on the growth performance of broiler chicken

0-28 day	Diet 1	Diet 2	Diet 3	Diet 4	Diet 5	SEM	*P-value*
Number of birds	60.00	60.00	60.00	60.00	60.00	-	
Initial body weight (grams/bird)	46.10	45.80	45.60	45.50	46.00	0.51	0.26
final body weight (grams/bird)	912.5^{b}	1120.7^{a}	1120.9^{a}	1130.1^{a}	1130.9^{a}	29.81	0.38
Body weight gain (grams/bird)	866.4^{b}	1074.9^{a}	1075.3^{a}	1084.6^{a}	1084.9^{a}	27.26	0.61
Average daily weight gain (grams/bird)	30.94^{b}	38.39^{a}	38.40^{a}	38.74^{a}	38.75^{a}	0.41	0.19
Total feed intake (grams/bird)	1300.3	1300.5	1300.6	1300.8	1300.9	31.83	0.37
Average daily feed intake (grams/bird)	46.44	46.45	46.45	46.45	46.46	0.50	0.25
Feed conversion ratio	1.50^{a}	1.21^{b}	1.20^{b}	1.20^{b}	1.20^{b}	0.07	0.03
Mortality (percentage)	1.50^{a}	1.00^{b}	0	0	0	0.02	0.01
29-56 days							
Body weight gain (grams/bird)	1902.7^{c}	2300.8^{b}	2319.5^{b}	2509.8^{a}	2600.1^{a}	51.42	0.02

Average daily weight gain (grams/bird)	67.95^{c}	82.17^{b}	82.83^{b}	90.64^{a}	92.86^{a}	0.72	0.06
Total feed intake (grams/bird)	3410.8^{c}	3610.2^{b}	3650.8^{a}	3655.1^{a}	3658.1^{a}	61.21	0.08
Average daily feed intake (grams/bird)	121.8^{c}	128.9^{b}	130.4^{a}	130.5^{a}	130.6^{a}	0.62	0.01
feed conversion ratio	1.80^{a}	1.57^{b}	1.57^{b}	1.46^{c}	1.41^{d}	0.06	0.01
Mortality (percentage)	1.00	0	0	0	0	0.02	0.01
Overall production cycle (0-56 days)							
Body weight gain (grams/bird)	1948.8^{c}	2346.6^{b}	2365.1^{b}	2555.3^{a}	2646.1^{a}	49.82	0.06
Average daily weight gain (grams/bird)	34.80^{c}	42.00^{b}	42.23^{b}	46.00^{a}	47.25^{a}	0.42	0.30
Total feed intake (grams/bird)	4711.1^{b}	4910.7^{a}	4951.4^{a}	4955.9^{a}	4959^{a}	105.2	0.03
Average daily feed intake (grams/bird)	84.13^{b}	87.69^{a}	88.42^{a}	88.50^{a}	88.55^{a}	0.70	0.06
Feed conversion ratio	2.42^{a}	2.09^{b}	2.09^{b}	1.94^{c}	1.87^{d}	0.03	0.04
Total mortality (percentage)	2.50^{a}	1.00^{b}	0	0	0		

Means on the same row having different superscripts are significantly different (P<0.05); SEM: Standard Error of Mean; Diet 1: basal diet without Muntinda calubara; diet 2: basal diet with 0.3 mL Muntinda calubara leaf extract/litre of water; diet 3: basal diet with 0.6 mL Muntinda calubara leaf extract/litre of water; diet 4: basal diet with 0.9 mL Muntinda calubara leaf extract/litre of water; diet 5: basal diet with 1.2 mL Muntinda calubara leaf extract/litre of water

Table 4. Effect of *Muntingia calabura* leaf extract on the carcass characteristics of broilers

Variables	Diet 1	Diet 2	Diet 3	Diet 4	Diet 5	SEM	*P-value*
Slaughter weight (grams)	1971.2^{c}	2435.7^{b}	2441.5^{b}	2804.1^{a}	2806.3^{a}	28.22	0.42
Carcass weight (grams	1421.5^{c}	1835.6^{b}	1821.9^{b}	2304.3^{a}	2306.5^{a}	23.97	0.30
Dressing percentage	72.11^{c}	75.36^{b}	74.62^{b}	82.18^{a}	82.19^{a}	0.90	0.02
Head (grams)	8.02	8.06	8.40	8.38	8.51	0.05	0.01
Legs (grams)	9.76^{c}	10.51^{b}	10.60^{b}	12.22^{a}	12.96^{a}	0.07	0.02
Breast (grams)	411.7^{c}	563.1^{b}	591.0^{b}	709.8^{a}	715.1^{a}	9.11	0.18
Thigh (grams)	310.0^{b}	314.1^{b}	306.3^{b}	490.1^{a}	495.7^{a}	4.38	0.21
Back (grams)	183.2^{c}	203.6^{b}	200.0^{b}	259.2^{a}	261.4^{a}	4.01	0.08
Wing (grams)	79.0	78.8	79.5	80.8	82.1	0.72	0.01
Drumstick (grams)	51.3^{c}	63.5^{b}	65.2^{b}	71.6^{a}	72.9^{a}	0.65	0.02
Liver (grams)	31.2	33.5	32.8	33.1	33.0	0.45	0.04
Gizzard (grams)	30.6^{c}	38.7^{b}	39.5^{b}	43.9^{a}	45.1^{a}	0.08	0.03
Heart (grams)	12.6	12.9	13.2	13.8	13.1	0.02	0.04

Means on the same row having different superscripts are significantly different (P<0.05); SEM: Standard Error of Mean; Diet 1: basal diet without Muntinda calubara; diet 2: basal diet with 0.3 mL Muntinda calubara leaf extract/litre of water; diet 3: basal diet with 0.6 mL Muntinda calubara leaf extract/litre of water; diet 4: basal diet with 0.9 mL Muntinda calubara leaf extract/litre of water; diet 5: basal diet with 1.2 mL Muntinda calubara leaf extract/litre of water

Table 5. Effect of *Muntingia calabura* leaf extract on the fatty acid of breast muscle of broilers

Variables	Diet 1	Diet 2	Diet 3	Diet 4	Diet 5	SEM	*P-value*
Saturated fatty acids							
Myristic acid	3.81^{a}	2.90^{b}	2.61^{b}	2.56^{b}	2.50^{b}	0.82	0.04
Palmitic acid	3.00^{a}	2.71^{b}	2.62^{b}	2.31^{b}	2.26^{b}	0.19	0.33
Strearic acid	13.81^{a}	13.74^{b}	13.83^{b}	10.91^{c}	10.02^{c}	0.21	0.10
Arachidic acid	5.55	5.45	5.22	5.06	5.9	0.23	0.11
Behenic acid	2.71^{a}	2.60^{b}	2.22^{b}	1.87^{c}	1.82^{c}	0.30	0.40
Lignoceric acid	0.92^{a}	0.71^{b}	0.69^{b}	0.44^{c}	0.40^{c}	0.18	0.50
Monounsaturated fatty acid							
Palmitoleic acid	2.16^{c}	3.88^{b}	3.73^{b}	5.11^{a}	5.17^{a}	0.66	0.04
Oleic acid	2.25^{c}	3.00^{b}	3.03^{b}	3.72^{a}	3.80^{a}	0.15	0.06
Erucic acid	10.64^{b}	10.22^{b}	10.70^{b}	12.02^{a}	12.71^{a}	0.35	0.01
Gadoleic acid	4.26^{c}	5.00^{b}	5.06^{b}	7.04^{b}	7.18^{a}	0.27	0.12
Nervonic acid	1.55^{c}	2.20^{b}	2.33^{b}	2.80^{a}	2.87^{a}	0.01	0.05
Polyunsaturated fatty acids							
Alpha-linolenic acid	10.22^{c}	15.00^{b}	15.27^{b}	20.09^{a}	20.11^{a}	0.09	0.40
Gamma linolenic acid	0.87^{c}	1.90^{b}	1.92^{b}	2.80^{a}	2.88^{a}	0.52	0.00
Arachidonic acid	6.11^{c}	9.30^{b}	9.38^{b}	12.41^{a}	10.56^{a}	0.06	0.01
Eicosadienoic acid	0.55^{c}	0.90^{b}	0.93^{b}	1.91^{a}	1.95^{a}	0.28	0.00
Docosapentaenoic acid	1.97^{c}	2.46^{b}	2.57^{b}	3.21^{a}	3.84^{a}	0.23	0.10
Decosatetraenoic acid	1.25^{c}	2.00^{b}	2.05^{b}	2.58^{a}	2.67^{a}	0.13	0.00
Total saturated fatty acid (%)	29.8^{a}	28.11^{a}	27.19^{b}	23.15^{c}	22.9^{c}	0.96	0.04
Total unsaturated fatty acid (%)	41.83^{c}	55.94^{b}	56.97^{b}	73.69^{a}	73.74^{a}	1.83	0.02

Means on the same row having different superscripts are significantly different (P<0.05); SEM: Standard Error of Mean; Diet 1: basal diet without Muntinda calubara; diet 2: basal diet with 0.3 mL Muntinda calubara leaf extract/litre of water; diet 3: basal diet with 0.6 mL Muntinda calubara leaf extract/litre of water; diet 4: basal diet with 0.9 mL Muntinda calubara leaf extract/litre of water; diet 5: basal diet with 1.2 mL Muntinda calubara leaf extract/litre of water

Table 6. Effect of Muntingia calabura leaf extract on the Immunoglobulins of broilers

Variables	Diet 1	Diet 2	Diet 3	Diet 4	Diet 5	SEM	*P-value*
Immunoglobulin A (µg/mL)	7.66^{c}	9.06^{b}	9.33^{b}	10.10^{a}	10.26^{a}	0.96	0.51
Immunoglobulin G (µg/mL)	2.29^{b}	3.31^{a}	3.38^{a}	4.05^{a}	4.15^{a}	0.05	0.04
Immunoglobulin M (µg/mL)	1.93^{b}	2.72^{a}	2.79^{a}	2.95^{a}	2.98^{a}	0.07	0.01

Means on the same row having different superscripts are significantly different (P<0.05); SEM: Standard Error of Mean; Diet 1: basal diet without Muntinda calubara; diet 2: basal diet with 0.3 mL Muntinda

calubara leaf extract/litre of water; diet 3: basal diet with 0.6 mL Muntinda calubara leaf extract/litre of water; diet 4: basal diet with 0.9 mL Muntinda calubara leaf extract/litre of water; diet 5: basal diet with 1.2 mL Muntinda calubara leaf extract/litre of water

3. Discussion

In the overall production cycle, the higher average daily weight gain was highest among birds fed diet 4 (0.9 mL *Muntingia calabura* leaf extract per liter of water) and diet 5 (1.2 mL *Muntingia calabura* leaf extract per liter of water) can be attributed to the presence of phyto-constituents in *Muntingia calabura* leaf extract as presented in Table 2. According to this study, *Muntingia calabura* leaf extract contains phenolic compounds, terpenoids, flavonoids, alkaloids, saponins and tannins at different concentrations, these compounds have been reported to possess numerous medicinal or herbal properties (Singh *et al*., 2021; Adewale *et al*., 2021). This results confirms the earlier study of Mahood et al. (2014), Mohini et al. (2021) who reveals that *Muntingia calabura* leaves contains flavonoids, alkaloids, tannins and phenols which have antimicrobial, anti-inflammatory, antioxidant, antifungal, antiviral, cytotoxic, anti-helminthic, analgesics, anti-diuretic, amongst others (Shittu *et al*., 2021; Musa *et al*., 2021; Agubosi *et al*., 2022). A synergistic combination of these phyto-constituents especially when administered at 0.9 mL (diet 4) and 1.2 mL (diet 5) per liter of water was capable of stimulating the secretion of digestive enzymes and saliva to improve the overall nutrient digestibility in birds (John, 2024; Alagbe, 2024). The antimicrobial properties in *Muntingia calabura* leaves also explains the reason why mortality was not recorded among birds fed the diet 3, 4 and 5. The presence of phenols, which is the most abundant phyto-constituents in this study could strengthen the gut epithelial barrier, thereby making it less prone to pathogenic organisms (Liz, 2020). Best feed conversion ratio was recorded among birds fed diet 3 and 4 relative to the other groups. The result recorded in this experiment is in consonance with the reports of Alqour et al. (2015) when plant extracts derived from thyme was fed to broilers at 2 mL/litre of water. Kanduri et al. (2013, Olujimi (2024) recorded similar results when phytogenics were fed to broiler chicken. Conversely, Oloruntola et al. (2021) recorded a non-significant difference in the feed intake of broilers fed Ocimum gratissimum leaf powder, this discrepancy could be attributed to the levels supplemented in the diet as well as the nature of bioactive compound in the test ingredient (John *et al*., 2024).

The highest dressing percentage were found in birds fed 0.9 mL and 1.2 mL *Muntingia calabura* leaf extract per liter of water in diet 4 and 5, intermediate in 0.3 mL and 0.6 mL *Muntingia calabura* leaf extract per liter of water and lowest in diet 1 (without *Muntingia calabura* leaf extract). Oloruntola et al. (2021). The current results are in agreement with the previous finding of Oloruntola et al. (2021) who reported that dressing percentage increased when broilers were fed 2.5 g/kg Ocimum gratissmum leaf powder as compared to the control. Similarly, Durrani et al. (2006) reported that dressing percentages were significantly increased when 0.5, 1.0 and 1.5% turmeric powder was included in the diets of broiler chickens. In disagreement with this current experiment, El-Ghamry (2002), supplemented Nigella sativa seeds and fresh garlic at 1.5g/kg and it revealed no significant changes in carcass weight and dressing percentage. Weights of head, wing, liver and heart were not significantly ($P>0.05$) influenced by the treatment except for those of legs, breast, thigh, back and drumstick ($P>0.05$), this results removes the possibility of toxicity from *Muntingia calabura* leaf extract since organs were not enlarged (Elagib *et al*., 2013). The increase in thigh,

breast, gizzard and breast could be due to the presence of the phyto-constituents in the test ingredient (Duru *et al*., 2013). Gizzard mechanically grinds food and acts as a microbial barrier due to its low pH and an increase suggests an improvement in digestive or metabolic capacity of birds (Mushtaq *et al*., 2014).

Meat quality can be influenced by many factors such as management, genetics, environment and nutrition (Diana, 2021; Alagbe *et al*., 2022). Total saturated fatty acid and total unsaturated fatty acid took the form of 29.80 – 22.90 % and 41.83 – 73.74 % accordingly was within the range reported by Kumar et al. (2015) when natural antioxidant was supplemented in the diet of broiler chicken. Similar outcome was observed by Goliomytis et al. (2014) when quercertin was supplemented in the diet of broilers. Unsaturated fatty acid (monosaturated fatty acid and polyunsaturated fatty acid) decrease the risk of heart disease by reducing the amount of low density lipoprotein (bad cholesterol) [LDL] and increasing high density lipoprotein [HDL]. Conversely, saturated fatty acid increases both LDL and HDL (Alagbe *et al*., 2022). Result obtained revealed that birds fed *Muntingia calabura* leaf extract had higher concentrations of unsaturated fatty acid and low saturated fatty acid relative to the control suggesting that the test ingredient has a tendency to lower cholesterol (hypolipidemic) in meat thus promoting food safety and increasing consumer's acceptability (Oloruntola *et al*., 2021). This is made possible by the presence of phytochemicals such as flavonoids and phenols in *Muntingia calabura* leaf extract. According to Alagbe (2021), flavonoids and phenols are capable of reducing the risk of hyperlipidemia in the serum and meat of animals.

Results on the effect of *Muntingia calabura* leaf extract on the Immunoglobulins of broilers showed that it influenced the activities of immunoglobin A (IgA), IgG and IgM in birds compared to those given control diet. The results suggest that *Muntingia calabura* leaf extract possess immune-modulatory properties thus reducing the exposure of birds to pathogenic attack. According to Mohini et al. (2021), leaves of Muntingia calabura contains bioactive compounds like; 20,40- dihydroxy chalcone (isoliquiritigenin (cabreuvin), (2S)-50-hydroxy-7,8,30,40-tetramethoxyflavan 20,40-dihydroxydihydrochal-cone and 3,4,5-trihydroxybenzoic acid which are strong antioxidants capable of producing antibodies in the system of birds. Immunoglobulin G has been reported to facilitate the process of phargocytosis and provides immunity to the system of animals while IgA and IgM are capable of protecting the mucosal membrane and blood stream to limit inflammation in the body (Agubosi *et al*., 2022). Result obtained in this study is in agreement with the reports of Begum et al. (2014) who observed an increase in the immunoglobulins of broilers fed herb mixture consisting of *Zingiber officinale*, *Platycordi radix* and *Phlomis umbrosa*.

4. Conclusion

In conclusion, *Muntingia calabura* leaf extract have beneficial effects on birds because they possess phyto-constituents that has antimicrobial, anti-inflammatory, sensorial stimulation, flavoring, immunostimulatory properties amongst others. Supplementing *Muntingia calabura* leaf extract at 0.9 mL and 1.2 mL per litre of water reveals that it can optimize the performance of birds without compromising the meat quality as well as the health status of animals. This research will further help to reduce the increasing rate of multidrug resistance and promote food safety.

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