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Effects of *Dichrostachys Glomerata* Feeding Regimes on Growth Performance, Gut Microbiota and Haemato-Biochemical Profile of Japanese Quails

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Abstract

Background: The ban of antibiotics growth promoters due to bacteria resistance and the presence of chemical residues in animal products have stimulated research for alternative feeding supplements in poultry production. This study was designed to assess the production performance of quails under two *D. glomerata* feeding regimes.

Methods: A total of 160 two weeks old Japanese quail chicks were assigned to four experimental treatments in a completely randomised design with 4 replicates of 10 chicks (5 males and 5 females) in each treatment. The two feeding regimes consisted of 5 g/kg of feed (T1) and cold water inclusion of 5 g/l (T2) of *D. glomerata*. Data were recorded on feed intake, weight gain, feed conversion ratio, haematological and serum biochemical parameters, and intestinal microbial count.

Findings: Quails fed on the two feeding regimes were compared to quails fed with diet without any supplement (T0) and an antibiotic (1 g/kg) medicated diet. The results showed no significant difference ($p>0.05$) in feed intake and weight gain, however the feed conversion ratio was significantly lower ($p<0.05$) with antibiotic compared to the two feeding regimes. The feeding regimes did not have any significant ($p>0.05$) effect on carcass yield and relative weight of organs. Triglycerides concentration was significantly higher in quails fed on cold water administration of *D. glomerata* as compared to the control diet. Aspartate aminotransferase (ASAT), alanine aminotransferase (ALAT), creatinine, total protein, albumin, urea, total cholesterol, HDL-cholesterol and LDL-cholesterol were not significantly affected by the feeding regimes. Except for the concentration of pack cell volume (PCV) that was significantly high ($p<0.05$) with cold water administration (41.5%) as compared to the negative control ration (35.25%), haematological blood components were not significantly affected by the feeding regimes. Feeding quails with *D. glomerata* powder whatever the regime significantly ($p<0.05$) increased lactic bacteria count compared to *E. coli*, *Salmonella* and *Staphylococci*.

Conclusions: The feeding of *D. glomerata* powder to quails through feed or drinking water can be used as an alternative to antibiotics to balance gut microbiota in Japanese quails.

Keywords *Dichrostachys glomerata*; Gut microbiota; Haemato-biochemical profile; Japanese quails; Spices

Introduction

Growth promoters like chemical products, herbal plants, essential oils, antibiotics and enzymes play an active role in the experimental and commercial production of large and small animals [1-2]. Antibiotics were added to animal feed in order to prevent, treat infections, reduce morbidity and mortality, as well as improve growth and production in animals [3]. Several studies showed that up to 81% of poultry meat and environmental isolates analysed were resistant to enrofloxacin, ciprofloxacin, tetracycline or erythromycin [4]. Sengül et al. [5] reported that dietary inclusion of flavomycin in quail's nutrition resulted in DNA damage and increased oxidative stress.

This resulted to the systematic removal of antibiotics from animal feed in the European Union countries [6]. The banning of antibiotic growth promoters and the increasing awareness of consumers triggered the need for potential alternatives to antibiotics among plant products which have been used for centuries such as spices and medicines.

Herbs and spices fall into the class of feed additives, currently referred to as "Phytogenics". They are strongly being considered as addition to the set of non-antibiotic growth promoters, such as organic acids and probiotics which are already well established in animal nutrition [7]. Some studies have shown that spices contain active substances which have a positive impact on the production performance of domestic animals [8-11]. The most important bioactive constituents include alkaloids, tannins, flavonoids, saponins and phenolic compounds.

D. glomerata has been shown to have antiviral, anti-infectious [12], anti-inflammatory and analgesic effects [13]. This fruit also exhibit *in vitro* and *in vivo* antioxidant activity and can inhibit oxidation of low-density lipoproteins [14]. These authors also reported the ability of *D. glomerata* to reduce fasting blood glucose and glycosylated hemoglobin levels.

This spices acts indirectly on broiler performance and meat quality characteristics through their antimicrobial, antioxidant and regulatory effects on animal's intestinal microflora [8,11,15]. It has inhibitory effects on *Staphylococcus epidermidis*, *Streptococcus viridans* and *Escherichia coli* [16].

The objective of this study was to evaluate the effects of *D. glomerata* feeding regimes on growth performance, gut microbiota and haemato-biochemical profile of Japanese quails.

Materials and Methods

Trial has been performed in agreement with the guidelines of the ethical standards from the 1964 Helsinki Declaration and latterly amendment.

Area of study

The study was carried out at the Teaching and Research Farm of the Faculty of Agronomy and Agricultural Sciences, University of Dschang, Cameroon. This farm is located at an altitude of 1420 m above sea level, between latitude 5°26′N and longitude 10°26′E with an equato-guinean climate, tempered by altitude.

Feed additives

Dried sample of *D. glomerata* fruit was bought at the local market in Dschang, ground into powder in a Harmed mill, sieved and incorporated in the experimental diets. The grounded spices sample was put in polyethylene plastic, sealed and stored at 4°C in a refrigerator until analysis.

Phytochemical screening of *D. glomerata* showed that tannin and alkaloids were absent. Steroids, flavonoids, saponins, anthraquinons, anthocyanines and triterpenoids tests were positive. Antibiotic (Doxycyclin®) used in the positive control diet was bought from a local veterinary pharmacy.

Animal and experimental diets

A total of 160 two weeks old Japanese quail chicks were randomly assigned to four treatments groups in a completely randomised design with 40 quails per treatment. Each group was sub divided into 4 replicates of 10 quails each (5 males and 5 females).

Two experimental diets were formulated from the negative control ration (T0) formulated to meet their requirements (**Table 1**) by incorporating 1 g of antibiotic/kg of feed and considered as positive control ration (T0+) and 5 g of *D. glomerata* fruit/kg of feed (T1). The last treatment consisted of incorporating 5 g/l of *D. glomerata* powder in drinking water (T2). Throughout the experiment, the quails received feed and water ad libitum every day.

Table 1: Composition of experimental diet

Ingredients (%)	Quantity (%)
Maize	60
Wheat brand	4.5
Soy bean meal	22
Fish meal	4.5
Bone meal	2
Oyster Shell	2
Premix 5%*	5
Total	100
Calculated nutrients composition	
Metabolisable energy (kcal/kg)	2906.8
Crude protein (%)	20.15
Calcium (%)	2.03
Phosphorus (%)	1.27
Lysine (%)	0.44
Methionine (%)	0.14

*Vitamin premix provided per kilogram of diet: Vitamin A: 3000000 IU; Vitamin D3: 600000 IU; Vitamin E: 4000 mg; Vitamin K: 500 mg; Vitamin B1: 200 mg; Vitamin B2: 1000 mg; Vitamin B6: 400 mg; Vitamin B12: 4 mg; Mn: 80 mg; Fe: 8000 mg; Zn: 10000 mg; Cu: 2000mg; Methionine: 200000 mg; Lysine: 78000 mg; Se: 20 mg.

Prophylactic measures against the most common infectious diseases were carried out. The quails were given treatment against coccidiosis (Vetacox®) and Vitamins (AMINTOTAL®) in water once a week. Chicks were weighed at the beginning of the experiment and on weekly basis thereafter. Data on feed intake and body weight gain were collected and used to calculate the feed conversion ratio (FCR) on weekly basis, monitored for five weeks.

Growth performance, haematological and serum biochemical parameters

At the end of the feeding trial (35 days of experiment), one male and one female from each replicate within the treatment (8 quails per treatment) were selected for carcass evaluation. They were fasted for 12 hours and slaughtered. Evisceration was done by hand plucking of feathers in warm water, removal of shank and viscera were recorded individually and presented as a percentage of live body weight.

From each slaughtered quail, blood was collected in 2 test tubes, one of which contained an anticoagulant. Blood with anticoagulant was used for haematological analysis using a fully automatic blood cell counter (Model PCE-21ON Hong Kong, China). Haematological parameters included White blood cell (WBC), Red blood cell (RBC), Haemoglobin (Hgb), Mean cell haemoglobin concentration (MCHC), Mean cell volume (MCV),

Mean cell haemoglobin (MCH) and Packed cell volume (PCV). Meanwhile, after centrifugation of blood free from anticoagulant, serum was collected and preserved at -20°C for the evaluation of total protein, albumin, globulin, aspartate aminotransferase (ASAT), alanine aminotransferase (ALAT), total cholesterol, high density lipoproteins (HDL), low density lipoproteins (LDL), triglyceride, urea and creatinine, using colorimetric method as prescribed by the commercial kits (Dialab® kits).

Microbial count

A sterile spatula was used to collect freshly quail droppings into sterile bottles. The identification and quantification of bacteria were assessed on appropriate specific culture media (MRS Agar for lactic acid bacteria, Mac Conkey AGAR for *E. coli*, SS AGAR for *Salmonella* and MST AGAR for *Staphylococcus*). Samples were incubated at 37°C for 24 hours.

Statistical analysis

Data recorded on growth, haematological, biochemical and microbial parameters were submitted to one-way analysis of variance test by General Linear Model procedure of Statistical Package for Social Science (SPSS 20.0) software. The differences were tested using Duncan's multiple range test and probability values less than 0.05 was considered as significant [17].

Results

Growth performance and carcass characteristics of grower Japanese quails

Table 2 summarised the overall performance of grower quails fed on *D. glomerata* feeding regimes. The results of the present study indicated no significant effects ($p>0.05$) among the treatment groups for feed intake, final body weight, and weekly weight gain.

However, the administration of *D. glomerata* powder through water (T2) numerically increased feed intake for about 2.89% and 8.37% compared to the negative (T0) and positive (T0+) control rations respectively. Water inclusion of *D. glomerata* significantly increased FCR compared to positive control ration supplemented with antibiotic.

Table 2: Growth performance and carcass characteristics of quails as affected by *D. glomerata* feeding regimes

	Treatments					
Growth parameters	**T0**	**T0+**	**T1 (Feed)**	**T2 (Water)**	**SEM**	**p-value**
Feed intake, g	148.37	139.99	141.93	152.78	6.03	0.15
Body weight, g	266.27	267.38	270.87	268.5	5.96	0.88
Weekly weight gain, g	35.07	34.4	34.16	34.57	1.25	0.9
FCR	4.23^{ab}	4.06^{b}	4.17^{ab}	4.43^{a}	0.15	0.02
Carcass characteristics (%BW)						
Carcass yield, %	77.69	78.85	77.79	77.59	0.25	0.25
Liver	1.71	1.72	1.81	1.66	0.04	0.54
Heart	1.06	0.97	0.99	0.97	0.03	0.69
Head	4.59	4.47	4.54	4.29	0.05	0.22
Legs	1.68	1.78	1.68	1.75	0.04	0.75
Abdominal fat	0.83	0.67	2.23	0.97	0.39	0.51

T0 =Negative control diet; T0+= T0 + 0.1% of Doxycycline; T1= T0 + 5 g *D. glomerata* powder/kg of feed; T2= drinking water + 5 g *D. glomerata* powder/litre of water.

Table 3 summarised the development of digestive organs of quails as affected by *D. glomerata* inclusion in feed and water. *D. glomerata* feeding regimes had no significant ($p>0.05$) effect on the development of digestive organs with respect to the negative and positive control treatments.

The haematological parameters of Japanese quails as affected by *D. glomerata* feeding regimes are summarised in **Table 4**. Although not significantly ($p>0.05$) different, feeding quails with *D. glomerata* whatever the regime tend to increase WBC, Hgb and MCH compared to negative and positive control ration. The PCV was significantly ($p<0.05$) higher with drinking water administration of *D. glomerata* compared to the negative control ration (T0).

Except for serum content in triglycerides which significantly increased ($p<0.05$) for about 20.62% in quails fed on *D. glomerata* inclusion in drinking water (T2) compared to the negative control treatment (T0), the concentration of serum biochemical parameters of grower Japanese quails after 5 weeks

of experimentation indicated no significant ($p>0.05$) difference among the serum constituents (**Table 5**).

Table 3: Effects of *D. glomerata* feeding regimes on the development of digestive organs of Japanese quails

	Treatments					
Digestive organs	**T0**	**T0+**	**T1 (Feed)**	**T2 (Water)**	**SEM**	**p-value**
Pancreas, %	0.2	0.2	0.2	0.19	0.01	0.95
Gizzard, %	1.89	1.83	2.04	1.89	0.05	0.4
Intestinal weight, g	5.16	5.16	5.18	5.6	0.15	0.71
Intestinal length, cm	60.98	62.43	62	64.61	0.9	0.56
Intestinal density, g/cm	0.08	0.08	0.08	0.09	0	0.85

T0 =Negative control diet; T0+= T0 + 0.1% of Doxycycline; T1= T0 + 5 g *D. glomerata* powder/kg of feed; T2= drinking water + 5 g *D. glomerata* powder/litre of water.

Table 4: Haematological parameters of Japanese quails fed on *D. glomerata*

	Treatments					
Blood parameters	**T0**	**T0+**	**T1 (Feed)**	**T2 (Water)**	**SEM**	**p-value**
WBC (103/ul)	83.73	84.1	85.95	85.43		0.39
RBC (106/ul)	3.6	3.57	3.69	3.83	0.06	0.41
Hgb (g/dl)	15.3	15.35	16.33	16.68	0.27	0.16
MCV (fl)	146.63	145.73	147.68	146.08	1.59	0.16
MCH (pg)	42.55	42.98	44.15	43.5	0.35	0.44
MCHC (g/dl)	29.03	29.48	29.9	29.78	0.18	0.34
PVC (%)	35.25^{b}	38.50^{ab}	37.50^{ab}	41.50^{a}	2.17	0.01

^{ab}: Means on the same row with different superscripts are significantly different ($p<0.05$); WBC= White blood cell; RBC= Red blood cell; Hgb= Haemoglobin; MCV= Mean cell volume; MCH= Mean cell haemoglobin; MCHC= Mean cell haemoglobin concentration; PCV= Packed cell volume; SEM= standard error on mean; T0 =Negative control diet; T0+= T0 + 0.1% of Doxycycline; T1= T0 + 5 g *D. glomerata* powder/kg of feed; T2= drinking water + 5 g *D. glomerata* powder/litre of water.

Table 5: Serum biochemical parameters of Japanese quails as affected by *D. glomerata* feeding regimes

	Treatments					
Biochemical parameters	**T0**	**T0+**	**T1 (Feed)**	**T2 (Water)**	**SEM**	**p-value**
ASAT (U/l)	274.27	319.13	285.81	296.19	17.28	0.84
ALAT (U/l)	46.34	58.4	42.42	79.7	7.79	0.34
Creatinine (mg/dl)	0.28	0.2	0.34	0.41	0.04	0.28
Total protein (mg/dl)	2.92	2.99	2.99	3.28	0.07	0.3
Abulmin (mg/dl)	1.65	1.83	1.83	1.52	0.1	0.54
Urea (mg/dl)	6.86	7.82	6.05	6.91	0.38	0.45
Total cholesterol (mg/dl)	153.07	159.18	173.01	158.83	9.25	0.9
HDL (mg/dl)	48.37	43.68	37.24	43.18	3.3	0.72
LDL (mg/dl)	88.91	101.18	113.67	95.87	9.02	0.82
Triglycerides (mg/dl)	93.49^{b}	109.17^{ab}	99.79^{ab}	117.78^{a}	4.02	0.15

^{ab}: Means on the same row with different superscripts are significantly different ($p<0.05$); SEM= standard error mean. T0= negative control diet; T0+= T0 + 0.1% of Doxycycline; T1= T0 + 5 g *D. glomerata* powder/kg of feed; T2= drinking water + 5 g *D. glomerata* powder/litre of water.

However, although not significant (p>0.05), the serum content in ASAT, ALAT and creatinine of quails fed on *D. glomerata* through drinking water numerically increased for about 7.40%, 41.86% and 31.70% respectively compared to quails fed on diet without supplement (T0).

The microbes isolated from the faecal sample of growing quails fed on *D. glomerata* are shown in **Table 6**. *E. coli* and *Salmonella* counts markedly decreased with antibiotic and *D. glomerata* in feed and water compared to the negative control treatment. Feeding quails with *D. glomerata* through drinking water or feed resulted in a significant increase in number of *Staphylococcus* counts. The feeding regimes had no significant (p>0.05) effect on lactic bacteria counts compared to the negative and positive control treatments. With regard to the bacteria species and irrespective of the treatment, lactic bacteria counts were significantly (p<0.05) high compared to *E. coli*, *Salmonella* and *Staphylococcus* counts.

Table 6: Faecal microbial loads of quails fed on *D. glomerata*

	Treatments					
Bacteria load log_{10}(cfu)	**T0**	**T0+**	**T1 (Feed)**	**T2 (Water)**	**SEM**	**p-value**
Lactobacillus	2.67	2.74	2.69	2.72	0.04	0.96
E. coli	2.28^{a}	1.89^{b}	2.03^{b}	1.94^{b}	0.05	0.02
Salmonella	1.66^{a}	1.19^{b}	1.46^{ab}	1.21^{b}	0.08	0.01
Staphylococcus	0.34^{b}	0.44^{b}	1.07^{a}	1.33^{a}	0.12	0

^{ab}: Means on the same row with different superscripts are significantly different (p<0.05); SEM= standard error on mean; T0= negative control diet; T0+= T0 + 0.1% of Doxycycline; T1= T0 + 5 g *D. glomerata* powder/kg of feed; T2= drinking water + 5 g *D. glomerata* powder/liter of water.

Discussion

Feeding quails with *D. glomerata* through drinking water resulted in a non-significant increase in feed intake for about 2.89% compared to the quails fed on the negative control diet. This may be as a result of the small dose of *D. glomerata* introduced, higher concentration of anti-nutritional substances or strong odour of this spice that may have reduced feed intake. Hernandez et al. [18] reported that optimisation of feed intake with feed additives from plant origin is controversial and depends on the amount and duration of administration. This corroborates the findings of Kalio et al. [19] who attributed the depression in feed intake of Japanese quails fed graded levels of *Azadirachta indica* leaf meal to the poor palatability and probable presence of toxic or anti-nutritional factors inherent in the spice. The present result agrees with the findings of Nweze et al. [20] who reported that feeding African porridge fruit (*Tetrapleura tetraptera*) pod to broilers through fresh or boiled water and feed has no effect on feed intake. Likewise the incorporation of 2 g of *D. glomerata*/kg of feed did not improve broiler feed intake [11].

Final body weight and weight gain were intermediate for all treatment groups. This result is in close agreement with Lee et al. [21] and Jang et al. [22] who reported no significant effect on the weight gain and live body weight of broilers fed on a commercial feed additive containing thymol and cinnamaldehyde. The present results are in contrast to the study of Oleforuh-Okoleh et al. [10] which revealed that adding 50 ml ginger, garlic and a combination of ginger-garlic into drinking water of broilers improved final body weight and weight gain compared to the negative control treatment.

The feed conversion ratio (FCR) significantly decreased (p<0.05) with feed inclusion of antibiotic compared to water inclusion of *D. glomerata*. The decrease in FCR can be understood because of the decrease in feed intake of birds fed on antibiotics. FCR of birds receiving *D. glomerata* in feed and water was comparable with birds receiving the negative control diet. The non-significant change in FCR with the feeding regimes could be associated with the dose of *D. glomerata* administered in this study. Also, this might be due to the antioxidant activity of *D. glomerata* that stimulated protein synthesis by bird's enzymatic system with respect to the quantity of feed or water consumed. Moreover, Muneendra et al. [23] stated that spices change fatty acid composition which affects the surviving ability of pathogenous microorganisms and could increase the digestive potentials of beneficial microorganisms there by increasing the animal's ability to absorb and use more nutrients or stimulate its immune system [24]. This suggests that 5g of *D. Glomerata*/kg feed was not enough to stimulate the absorption and usage of more nutrients.

ANOVA revealed that *D. glomerata* powder incorporated in feed and water did not significantly affect carcass yield, liver, pancreas, heart, gizzard, head, legs and abdominal fat and relative weight of the intestine when compared to the negative and positive control treatments. This might be because of the small quantity, short duration of administration of *D. glomerata* in the diets, the small quantity of feed consumed and metabolised by quails. This result agrees with the results of Barad et al. [25] who reported that 2% spices of coriander seed, turmeric powder and 0.5% black pepper had no significant effect on broilers carcass yield and relative weight of organs. Moreover, Seifi et al. [26] reported that organic acid supplementation of broiler diet had no significant effect on the length of the small intestine or proportional weight of liver and pancreas. Also, Fazilat et al. [27] showed that weights of quail's organs like gizzard, heart, liver, kidney and spleen are not affected by the supplementation of ration with commercial feed additive (Globacid®). This contradicts the results of Simsek et al.

[28] who reported that, the addition of anise oil and essential oil mix (Herbomix®) to diets had positive effects on carcass yield in broilers. In addition, Goodarzi and Nanekarani [29] recorded significant effect only on abdominal fat when 1% and 2% onion extracts were introduced in the drinking water of broiler chickens.

In the present study, with the exception of PCV which was significantly ($p<0.05$) higher compared to the negative control treatment, all other haematological parameters of quails were not significant among treatments. Haematological parameters in feeding regimes had an upward trend compared to the negative control treatment. This might be due to the absence of toxic substances like alkaloids and tannins in the spice which did not provoke a counteraction of the immune system which could markedly decrease WBC, RBC, Hgb and PCV. Also, Oleforuh-Okoleh et al. [10] and Islam et al. [30] reported that an increase in RBC, Hgb and PCV contents in blood is an indication of improved oxygen carrying capacity of the cells which translates a better availability of nutrients to the birds consequently affecting their wellbeing. This corroborates the findings of Onu [31] who observed no significant effects on WBC, RBC, Hgb, MCV, MCH and MCHC when 0.25% garlic, ginger and a combination of garlic-ginger were incorporated in finisher broiler's feed. In addition, Parthasarathi et al. [32] recorded a marked increase in PCV and no significant effect on MCV and MCH when 1% ginger, garlic and equal combination of ginger-garlic powder were added in Japanese quail's feed. The present results are in contradiction with the findings of Al-Kassie et al. [33] who observed a significant decrease in WBC, RBC, Hgb and PCV when 0.25%, 0.5%, 0.75% and 1% hot red pepper was introduced in broiler's diet compared to the negative control diet. Furthermore, Oleforuh-Okoleh et al. [10] reported a significant increase in WBC, RBC and Hgb compared to the negative control treatment when 50 ml ginger, garlic and a combination of ginger-garlic were incorporated in broiler's drinking water.

The serum contents in ALAT (alanine amino-transferase) and ASAT (aspartate amino-transferase) were not significantly affected by the incorporation of *D. glomerata* powder in feed or water suggesting that *D. glomerata* had no negative effect on the liver. This might be due to the hepato-protectory effects of the substances present in the studied spice. This results agrees with the findings of Kana et al. [11] which revealed that, the incorporation of 0.2 and 0.4% *D. glomerata* in the diet of broilers did not significantly affect serum content in ASAT and ALAT. This observation contradicted the finding of Rehman et al. [34] who reported that feeding broiler with a mixture of aqueous extracts of medicinal plants induced a reduction in ALAT and ASAT ratios. This contradiction can be due to the multitude of the active compounds in the mixture of the extracts used by these authors which could have affected liver function.

Creatinine level had an upward trend with *D. glomerata* compared to the negative and positive control treatments. This might be as a result of the absence of bioactive substances like alkaloids and tannins in the spice that can be harmful to the kidney. This result agrees with the findings of Onu [31] and Al-Shuwaili et al. [35] who reported no marked effect on creatinine level when 0.25% garlic, ginger, combination of garlic-ginger and 5% spices of garlic, ginger, and cinnamon were respectively fed to broilers and turkeys. The present results contradicts the findings of Kana et al. [11, 36] who respectively reported a marked ($p<0.05$) increase in creatinine content with 0.4% *D. glomerata* and *Afrostyrax lepidophyllus* (bark and combination of fruit-bark) in broiler chickens.

The serum content in urea and total protein were not affected by the supplementation of feed and drinking water with *D. glomerata*. This suggests that the inclusion of *D. glomerata* fruit powder in quail's diet does not have harmful effects on kidney function (serum rate of urea) and the immune system (serum protein rate). This is in accordance with the findings of Al-Shuwaili et al. [35] which showed that incorporating ginger, garlic and cinnamon in turkey's ration had no marked effects on total protein level. This result contradicts the results of Ngouana et al. [37] which revealed that, the incorporation of essential oils encapsulated in chitosan and charcoal in broilers feed increased the serum content in total protein and serum urea levels.

The level of serum cholesterol, high density lipoproteins and low density lipoproteins were not affected by dietary treatments. This contradicts the results of Ali et al. [38] who reported that the addition of thyme in the diet of chicken induced a significant decrease in the serum content in HDL-cholesterol and total cholesterol. Meanwhile, feeding quails with *D. glomerata* inclusion in water markedly increased serum content in triglycerides compared to the negative control treatment. This could be explained by the ability of the spice to increase the secretion of bile salts which would have led to a better digestion of dietary lipids. This result is in agreement with the findings of Bolukbasi et al. [39] who reported that essential oil of thyme leads to an increase in the triglyceride level in broiler chickens.

The numbers of lactic bacteria count (beneficial bacteria) were higher than *E. coli*, *Salmonella* and *Staphylococcus* (pathogenic bacteria) counts. This could be explain by the presence of substances like phenols in spices which promote the development of lactic bacteria. Also, antimicrobial property of the spice is considered to arise from phenols, flavonoids [40] and hydrophobic essential oil present in spices and plants that intrude into the bacterial cell membrane, disintegrate the membrane structure and cause leakage thus making microbes less virulent [7,23]. More to that, phenolic compounds act by forming complexes with several proteins causing the destruction of microbe's membrane rendering unavailable certain substrates for microbes [40]. Moreover, Guo et al. [41] stated that spices and plant extracts leads to a decrease in the number of harmful microorganisms while at the same time increase the number of beneficial microorganisms. This result agrees with the findings of El-Shenway and Ali [42] who observed an increase in lactic bacteria population in the intestine of healthy quails. The suppression of harmful microorganisms (*E. coli* and *Salmonella*) resulted to better growth and metabolism of beneficial microbes although not significant. Antimicrobial activity of natural extracts and compressed oils are closely linked with their polyphenolic content [43]. Therefore, plants or their extracts

rich in phenolic and other bioactive compounds may serve as potential natural antimicrobial agents [44].

Staphylococcus increased significantly ($p<0.05$) in *D. glomerata* feeding regimes compared to the negative control treatment. This could be explain by the absence of substances like alkaloids and tannins that might have played a greater role in destroying the microorganism. Pavithra [45] reported that spices with alkaloids have good antimicrobial property against gram positive microorganisms like *S. aureus* and *S. typhi*. Also, Harris et al. [46] reported that for Staphylococci to proliferate easily, they must have access to their nutritional requirements generally made of an organic source of nitrogen supplied by 5-12 essential amino acids. These amino acids might have been obtained from quail's digestion of proteins present in their feed with respect to the quantity consumed thereby giving room for *Staphylococcus* proliferation. Furthermore, they are resistant microorganisms as they tolerate antibiotics, high temperatures and concentration of salts [47]. This could be due to the thick cell wall they possess which provides the organism with a great internal pressure making it nearly impossible for the antimicrobial spices or drugs to enter the cell [48]. These results contradicts the findings of Nassan et al. [49] who reported that *in vivo* and *in vitro* evaluation of clove water extract respectively in male albino rats and using agar well diffusion method had antimicrobial activity against *Staphylococcus aureus* at a minimum inhibitory concentration (MIC) of 2 mg/ml. This suggests that *D. glomerata* had a MIC less than what was required to considerably reduce Staphylococci population.

Conclusion

This study revealed that the administration of *D. glomerata* powder to quails through feed or drinking water is not significantly effective on growth performance. However, *D. glomerata* whatever the feeding regime can be used to balance gut microbiota and haemato-biochemical parameters of Japanese quails.

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