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Research Article
The Effect of Medicinal Herb Inclusion on Hematologic Status and Blood Lipid Profiles in Broiler Chickens

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Abstract

Background and Objective: The use of antibiotics as a feed supplement in broiler chickens is not recommended and therefore, antibiotics should be substituted with alternative compounds. The present study was conducted to evaluate the effect of medicinal herb powder inclusion on hematological status, blood lipid profiles, internal organ weight and toxicity score in female broiler chickens. **Materials and Methods:** One hundred sixty-eight female broiler chickens aged 15 days were distributed into 7 groups. Each treatment group consisted of 4 replicates and each replicate consisted of 6 broilers. The 7 treatments were as follows: 1) broilers were fed a diet with no medicinal herbs as the control (P0), 2) broilers were fed with 5% *Sauropus androgynus* (*S. androgynus*) leaf powder (P1), 3) broilers were fed a diet with 5% bay leaf powder (P2), 4) broilers were fed a diet with 5% basil leaf powder (P3), 5) broilers were fed a diet with 5% papaya leaf powder (P4), 6) broilers were fed a diet with 5% *Moringa* leaf powder (P5) and 7) broilers were fed a diet with 5% noni fruit powder. **Results:** *Sauropus androgynus* leaf contained the highest protein content, at 34.37%, while noni fruit and basil leaf were rich in iron, at 7.80 and 6.43 ppm, respectively, followed by papaya leaf and *Sauropus androgynus* leaf, at 5.61 and 4.02 ppm, respectively. The noni fruit contained the highest crude fiber content (19.33%) followed by bay leaf (17.73%) and *Sauropus androgynus* leaf (14.16%). Experimental results showed that medicinal herb inclusion affected WBC ($p<0.01$) and lymphocytes ($p<0.01$) but had no effect on RBC, Hb, PCV, MCV, MCH, MCHC and thrombocytes, P4 had the lowest WBC counts ($p<0.01$). Experimental results showed that the inclusion of medicinal herbs increased HDL ($p<0.01$), reduced LDL ($p<0.05$), triglycerides ($p<0.01$) and LDL/HDL ratio ($p<0.01$) but had no effect on total cholesterol. **Conclusion:** Selected medicinal herb inclusion reduced blood triglycerides, LDL and LDL/HDL ratio but increased HDL. Papaya and *Sauropus androgynus* leaves increased lymphocyte counts.

Key words: Medicinal herbs, hematologic status, blood lipid profiles, broiler chickens, toxicity score, metabolic secondary compounds, flavonoid fractions

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Competing Interest: The authors have declared that no competing interest exists.

Data Availability: All relevant data are within the paper and its supporting information files.

INTRODUCTION

The use of antibiotics as a feed supplement in broiler chickens is not recommended because antibiotics accumulate in the meat. Antibiotic residues cause pathogenic microbial resistance, however, antibiotics are needed in broiler chickens in order to produce optimal performance and meat quality. In addition, the poultry industry is faced with consumer pressure to produce meat that is low in cholesterol. This demand is confirmed by experimental results that show a high concentration of blood fat, such as cholesterol, which is correlated with the risk of atherosclerosis, coronary heart disease, stroke and other metabolic diseases[1].

Commercial feed additives contain only a small number of vitamins, micro-minerals, antioxidants and antibiotics. In general, commercial feed additives prepared with synthetic substances were shown to have more side-effects such as damaging hormonal and immune systems[2] and increasing blood lipid concentrations[3].

To overcome these problems, some herbs that have antioxidant properties have been suggested as a replacement for antibiotics[4]. Medicinal herbs have low side effects and are able to maintain the product quality and performance of livestock[5]. Antioxidant compounds found in medicinal plants include α-tocopherol (vitamin E), β-carotene, ascorbic acid, flavonoids, carotenoids, anthocyanins, phenolic compounds, zinc and selenium[6,7].

Some herbs that have the potential as alternative feed additives include *Sauropus androgynus* leaf[8-14], basil leaf[15-17], papaya leaf[18-19], noni fruit[20], bay leaf[21-22] and *Moringa* leaf[23-27].

Sauropus androgynus leaf contains 6 major compounds-monomethyl succinate and cis-2-methyl cyclopentanol acetate, benzoic acid, acid phenyl malonate, 2-pyrolidinon and methyl pyroglutamate[8] (converted to glutamate), succinate, acetate and estradiol-which have an important role in the metabolism of nutrients[3]. Santoso and Sartini[9] found t hat adding *Sauropus androgynus* leaf powder at 3% to broiler rations improved feed efficiency and decreased abdominal fat deposition and carcass fat content. *Sauropus androgynus* leaves contains β-carotene at 3510.3 mg g^{-1}. *Sauropus androgynus* leaves ground into powder are also rich in iron and amino acids[10] and have antioxidant and antibacterial properties[11-14].

Essential oils of basil leaves show antibacterial activity against *S. aureus* and *E. coli*[15,16]. Furthermore, the essential oil of basil leaves contain hydrocarbons, alcohols, esters, phenol (1-19% eugenol, iso-eugenol), phenolat ether (methyl clavicol 3-31%, 1-9% methyl eugenol), oxides and ketones. Increased body weight was caused by the improved hematologic profile of broiler chickens[17]. Santoso[18] reported that the supplementation of papaya leaf extract improves body weight gain and carcass quality in broiler chickens. Santoso and Fenita[19] reported that the administration of papaya leaf powder increases the protein content in eggs.

Fenita[20] reported that the administration of noni powder at 3% reduces the concentration of cholesterol and triglycerides in the blood to below 50%.

Bay leaves are rich in minerals and vitamins as well as essential oils[21]. Essential oils, saponins, flavonoids and tannins in the leaves have antibacterial properties against *Salmonella* sp., *Bacillus cereus*, *B. subtilis*, *Staphylococcos aureus*, *E. coli* and *Pseudomonas fluorescens*[22].

Moringa leaves present antioxidant, anticancer, anti-atherosclerotic, anti-inflammatory and antitumor properties, regulate thyroid status, improve the body weight gain of broilers and improve the immune system[23-26]. *Moringa* leaves contain phenolic compounds at levels of 115.68 mg GAE g^{-1} dry extract, flavonoids at levels of 113.95 mg QE g^{-1} dry extract and prevent autooxidation[27].

The present study was conducted to evaluate the effect of selected medicinal herbs on the hematologic status and blood lipid profiles in broiler chickens.

MATERIALS AND METHODS

Medicinal plant powders: Basil leaves, *Sauropus androgynus* leaves, bay leaves, noni fruit, papaya leaves and *Moringa* leaves obtained from the field were air-dried for 5 days. Afterwards, they were dried in the sun for 1 h so that the level of moisture was reduced to approximately 10-12%, then, they were milled and stored in plastic bags.

Broiler chickens, housing and feeding: Three hundred broiler chickens aged 1 day were placed in a cage broiler. Broiler new arrivals were given sugar water to reduce the stress of travel. Brooder temperature was set in accordance with the standards of maintenance procedures. At the age of 4 and 21 days, broiler chickens were vaccinated ND. At the age of 1-13 days, the broiler chickens were fed commercial diets.

At 14 days old, female broilers were selected and distributed into experimental plots and fed experimental diets until the age of 35 days. Experimental diets contained 19% crude protein and 3.200 kcal ME kg^{-1}. The feedstuff compositions used in the present study are presented in Table 1. Medicinal herb powder was used at a maximum of 5%[10,28].

One hundred sixty-eight female broiler chickens aged 15 days were distributed into 7 groups. Each treatment group consisted of 4 replicates and each replicate consisted of 6 broilers. The 7 treatments were as follows: (1) Broilers were

Table 1: Feedstuffs used in the present study

Feedstuffs (%)	P0	P1	P2	P3	P4	P5	P6
Yellow corn	57.0	55.0	55.0	55.0	55.0	55.0	55.0
Rice brans	5.0	4.0	5.0	5.0	5.0	5.0	5.0
Fish meal	7.0	7.0	7.0	7.0	7.0	7.0	7.0
Soybean meal	27.5	26.0	26.0	26.0	26.0	26.0	26.0
Palm oil	1.0	1.0	1.0	1.0	1.0	1.0	1.0
Mineral mixture	2.0	2.0	2.0	2.0	2.0	2.0	2.0
Top mix (commercial feed additive)	0.5	0	0	0	0	0	0
Sauropus leaf		5.0					
Bay leaf			5.0				
Basil leaf				5.0			
Papaya leaf					5.0		
Moringa leaf						5.0	
Noni fruit							5.0
Total	100	100	100	100	100	100	100
Calculated composition							
Protein (%)	20.12	20.91	19.80	20.57	20.35	20.58	19.74
Ca (%)	1.02	1.13	1.08	1.14	1.15	1.12	1.09
P (%)	0.73	0.75	0.72	0.76	0.73	0.74	0.72
Fat (%)	3.31	3.41	3.39	3.37	3.51	3.36	3.33
Ash (%)	4.20	4.46	4.16	4.58	4.51	4.40	4.21

P0 = Control, P1 = *Sauropus androgynus* leaf, P2 = Bay leaf, P3 = Basil leaf, P4 = Papaya leaf, P5 = *Moringa* leaf, P6 = Noni fruit

fed a diet without medicinal herbs as the control (P0), (2) Broilers were fed with 5% *Sauropus androgynus* leaf powder (P1), (3) broilers were fed a diet with 5% bay leaf powder (P2), (4) broilers were fed a diet with 5% basil leaf powder (P3), (5) broilers were fed a diet with 5% papaya leaf powder (P4), (6) broilers were fed a diet with 5% *Moringa* leaf powder (P5) and (7) broilers were fed a diet with 5% noni fruit powder.

Broiler chickens were maintained in accordance with the standards of maintenance procedures of broiler chickens and given diets and water *ad libitum.*

Sampling and laboratory analysis: At the end of the experiment (35 days of age), 4 broiler chickens in each treatment group were selected and blood samples were collected by puncturing wing veins using a 3 mL syringe and placed in sample bottles without anticoagulant agent[29]. Immediately after blood collection, the sample bottles were gently shaken to prevent lysing of the blood[30].

The packed cell volume (PVC) was determined by the micro-hematocrit method[31]. The hemoglobin was determined using the cyanmethemoglobin method, while the red blood count (RBC) was carried out using the hemocytometer method[32]. The thrombocyte count was determined using the Ress-Ecker method[33] and the white blood count (WBC) was determined using hemocytometer method[32].

To obtain the plasma, blood samples were collected, bottled with anticoagulant agent and centrifuged at 3000 rpm. Blood serums obtained were then analyzed for triglyceride, cholesterol, high-density lipoprotein (HDL) and low-density lipoprotein (LDL). Total cholesterol and LDL were determined by the method of Kulkarni[34] and triglyceride concentrations were determined by the method of Fossati and Prencipe[35].

Internal organs (liver, heart, intestine, gizzard, spleen) were removed and weighed and then calculated by the following equation[29]:

$$\text{Internal organ (\%)} = \frac{\text{Internal organ weight}}{\text{Live wight}} \times 100$$

Toxicity score was calculated by the following equation[29]:

$$\text{Toxicity (\%)} = \frac{\text{Liver+spleen weights}}{\text{Live wight}} \times 100$$

Statistical analysis: The results of the study were subjected to a one-way analysis of variance and if significantly different were tested further by Duncan's multiple range test. In all cases, differences were considered significant at $p<0.05$.

RESULTS

Herbal chemical composition: Table 2 presents the chemical composition of selected medicinal plants. *Sauropus androgynus* leaf contains the highest protein content, at 34.37%, while noni fruit and basil leaf were rich in iron, at 7.80 and 6.43 ppm, respectively, followed by papaya leaf and *Sauropus androgynus* leaf at 5.61 and 4.02 ppm, respectively. The noni fruit contains the highest crude fiber content (19.33%), followed by bay leaf (17.73%) and *Sauropus androgynus* leaf (14.16%).

Table 2: Chemical composition of selected medicinal herbs

Components	*Moringa* leaf	Noni fruit	Papaya leaf	*Sauropus* leaf	Basil leaf	Bay leaf
Moisture (%)	9.95	10.56	9.69	10.49	10.11	9.24
Ash (%)	8.85	5.14	11.11	10.13	12.56	4.16
Protein (%)	27.67	10.98	23.26	34.37	27.65	12.30
Fat (%)	3.70	3.15	6.66	4.78	4.01	4.27
Crude fiber (%)	5.31	19.33	8.85	14.16	5.59	17.73
Calcium (%)	2.09	1.48	2.72	2.38	2.46	1.30
Phosphor (%)	0.67	0.27	0.64	0.83	1.06	0.28
Iron (ppm)	2.00	7.80	5.61	4.02	6.43	1.59
Energy (kcal kg^{-1})	484.2	426.3	421.8	378.8	379.5	388.3

Table 3: Metabolic secondary of selected medicinal herbs

	Moringa leaf	Noni fruit	Papaya leaf	*Sauropus* leaf	Basil leaf	Bay leaf
Alkaloids	-	-	-	-	-	-
Flavonoids	+	+	+	+	+	+
Triterpenoids	+	+	+	+	+	+
Phenols	+	+	+	+	+	+
Hydroquinone	-	-	-	-	-	-
Saponins	+	-	+	+	-	+
Tannins	+	+	+	+	+	+

Table 4: Flavonoids fraction in selected medicinal herbs (mg/100 g)

	Moringa leaf	Noni fruit	Papaya leaf	*Sauropus* leaf	Basil leaf	Bay leaf
Myricetin	-	-	-	-	-	-
Luteolin	-	-	-	-	-	-
Quercetin	76.26	23.64	33.64	27,85	21.48	28.49
Apigenin	-	-	-	-	-	-
Kaempferol	9.45	2.48	8.97	2.26	1.74	4.36

Table 5: Effect of selected medicinal herbs on hematologic profiles in broiler chickens

Variables	P0	P1	P2	P3	P4	P5	P6
WBC (10^3 mL^{-1})	295.3±7.8^{b}	294.60±23.1^{b}	273.3±8.1^{b}	283.5±44.6^{b}	195.7±5.5^{a}	284.6±7.7^{b}	288.5±7.5^{b**}
RBC (10^6 mL^{-1})	2.87±0.17	3.14±0.16	2.80±0.59	2.96±0.18	2.69±0.27	2.67±0.07	2.80±0.08^{ns}
Hb (g dL^{-1})	11.33±0.17	11.90±0.88	11.33±2.42	10.78±0.82	12.75±1.14	10.73±0.43	1.75±0,19^{ns}
PCV (%)	36.5±0.58	38.75±2.63	36.50±7.68	35.75±2.22	34.50±3.11	34.00±0.82	34.25±0.508^{ns}
MCV (fl)	124.8±2.5	124.3±3.78	124.8±3.30	123.0±1.83	127.3±2.50	126.8±2.75	122.5±2.38^{ns}
MCH (pg)	37.50±1.29	37.00±0.82	37.00±0.00	36.50±0.58	37.25±0.96	38.50±1.73	38.25±0.96^{ns}
MCHC (g dL^{-1})	30.25±0.50	30.50±0.58	30.00±0.00	30.50±1.00	33.25±4.57	31.25±0.96	30.35±0.50^{ns}
Thrombocyte (10^3 μL^{-1})	2.75±0.96	4.25±0.50	4.00±0.82	3.00±0.82	3.25±1.26	3.25±0.96	2.50±0.58^{ns}
Lymphocyte (%)	0.95±0.01^{b}	0.96±0.02^{c}	0.95±0.01^{b}	0.94±0.01^{ab}	0.96±0.01^{c}	0.94±0.01^{ab}	0.93±0.01^{a**}

P0: Control, P1: *Sauropus androgynus* leaf, P2: Bay leaf, P3: Basil leaf, P4: Papaya leaf, P5: *Moringa* leaf, P6: Noni fruit

Table 3 shows the content of secondary metabolic compounds in selected medicinal plants. All plants contained flavonoids, triterpenoids, tannins and phenols. All selected medicinal plants did not contain alkaloids. Noni fruit and basil leaf did not contain saponins.

Table 4 presents the flavonoid fraction content in selected medicinal plants. All medicinal plants did not contain myricetin, luteolin and apigenin. *Moringa* leaf contained the highest level of quercetin, at 76.26 mg, while basil leaf contained the lowest, at 21.48 mg. *Moringa* leaf also contained the highest level of kaempferol, at 9.45 mg, while basil leaf had the lowest (1.74 mg).

Hematologic profiles: Table 5 shows the effect of selected medicinal herbs on the hematologic profile of broiler chickens. Experimental results show that medicinal herb inclusion affected WBC ($p<0.01$) and lymphocytes ($p<0.01$) but had no effect on RBC, Hb, hematocrit, MCV, MCH, MCHC and thrombocytes. P4 had the lowest WBC ($p<0.01$). P6 had the lowest lymphocyte count, whereas, P1 and P4 had the highest.

Internal organ weight and toxicity score: Table 6 shows the effect of selected medicinal herbs on internal organ weight and toxicity score. Experimental results showed that the included medicinal herbs had no effect on liver, spleen, gizzard, heart, intestine weight and toxicity scores.

Blood lipid profiles: Table 7 shows the effect of selected medicinal herbs on blood lipid profiles. Experimental results showed that the included medicinal herbs affected HDL

Table 6: Effect of medicinal herbs on internal organ weight and toxicity score

Variables	P0	P1	P2	P3	P4	P5	P6
Liver (%)	2.28±0.67	1.91±0.11	2.30±0.34	1.94±0.15	2.11±0.34	1.91±0.89	1.99±0.17^{ns}
Spleen (%)	0.15±0.11	0.09±0.01	0.16±0.08	0.09±0.01	0.16±0.10	0.10±0.01	0.08±0.02^{ns}
Gizzard (%)	1.68±0.16	1.60±0.30	1.75±0.16	2.07±0.23	2.06±0.26	1.97±0.50	1.62±0.21^{ns}
Heart (%)	0.33±0.05	0.32±0.04	0.35±0.03	0.33±0.04	0.35±0.05	0.31±0.01	0.32±0.03^{ns}
Intestine (%)	2.48±0.36	2.51±0.59	3.27±0.23	2.71±0.32	3.08±0.36	2.92±0.36	3.01±0.34^{ns}
Toxicity score	2.43±0.78	1.30±0.11	2.46±0.41	2.04±0.15	2.26±0.39	2.01±0.09	2.06±0.18^{ns}

P0: Control, P1: *Sauropus androgynus* leaf, P2: Bay leaf, P3: Basil leaf, P4: Papaya leaf, P5: *Moringa* leaf P6: Noni fruit

Table 7: Effect of medicinal herbs on blood lipid profiles

Variables	P0	P1	P2	P3	P4	P5	P6
Cholesterol (mg dL^{-1})	130.00±5.32	128.00±2.52	125.00±1.89	129.00±2.75	128.00±2.06	131.00±7.54	127.00±2.16^{ns}
HDL (mg dL^{-1})	36.00±0.50^{a}	39.00±1.87^{e}	38.00±1.67^{cde}	37.00±1.22^{abc}	38.00±1.14^{cde}	37.00±0.55^{ab}	39.00±1.10^{de**}
LDL (mg dL^{-1})	94.00±5.60^{bc}	87.75±3.10^{ab}	86.75±2.22^{a}	92.00±1.83^{abc}	89.50±1.91^{abc}	94.75±8.10^{c}	87.75±2.22^{ab*}
Triglyceride (mg dL^{-1})	40.50±3.11^{d}	37.00±2.16^{bc}	34.00±1.41^{a}	39.00±1.41^{cd}	36.25±0.50^{ab}	40.50±1.29^{d}	38.00±0.82^{bcd**}
LDL/HDL	2.60±0.18^{c}	2.21±0,12^{a}	2.29±0.14^{ab}	2.47±0.07^{bc}	2.35±0.10^{ab}	2.60±0.26^{c}	2.24±0,07^{ab**}

P0: Control, P1: *Sauropus androgynus* leaf, P2: Bay leaf, P3: Basil leaf, P4: Papaya leaf, P5: *Moringa* leaf, P6: Noni fruit

(p<0.01), LDL (p<0.05), triglyceride (p<0.01) and LDL/HDL ratios (p<0.01) but had no effect on total cholesterol. P0 had the lowest HDL, whereas, P1 had the highest. P0 and P5 had the highest LDL. P0 had the highest triglyceride, whereas, P2 had the lowest. P0 and P5 had the highest LDL/HDL ratio, whereas, P1 had the lowest.

DISCUSSION

The results of present study show that *Moringa* leaf contained 27.67% protein, 2.09% calcium, 0.67% phosphorus and 2 ppm iron. Halaby *et al.*[36] reported that *Moringa* leaf powder contained 27.81% protein. In comparison with the results of Halaby *et al.*[36], the present study showed higher calcium and phosphorus but lower iron levels. It means that *Moringa* leaf could be used as protein, calcium and phosphorus sources for broiler chickens.

The protein content of *Sauropus androgynus* leaf powder in this study was higher (34.37%) than was observed by Andarwulan *et al.*[37], who reported that this herb contained 25.73% protein. Basil leaf protein in the present study was higher (27.65%) than observed by Mlitan *et al.*[38], who reported that basil leaves had 9.10% protein. However, the present study showed lower fat, ash and iron contents compared to the observations of Mlitan *et al.*[38], who found that basil leaves contained 10.80% fat, 14.30% ash and 4.35 mg iron/100 g, The noni fruit protein content in the present study was lower than the observation of Lujan *et al.*[39], who reported that the protein content of fresh noni fruit ranged from 5.96-15.6%. Dev and Iqbal[40] reported that dried papaya leaf tea contains 4.7% moisture, 26.2% protein, 2.6% fat and 10.8% ash. Thus, the results of the present study showed that four medicinal herbs (*Sauropus androgynus* leaf, papaya leaf, *Moringa* leaf and basil leaf) could be used as protein sources for broiler chickens. Protein plays an important role in hematological status; for example, protein inclusion increases red blood cells, hemoglobin[41] and lymphocytes[42].

The present study showed that the selected medicinal herbs contained flavonoids, triterpenoids, phenols and tannins. *Sauropus androgynus* contained tannins, saponins, alkaloids, flavonoids, terpenoids, phenols and sterol[43], whereas, papaya leaf contained alkaloids, saponins and alkaloids but had no tannins[44]. Ojiako[45] reported that *Moringa* leaf contained tannins, alkaloids, saponin and phenol, whereas, Anjorin and Ugwu[46] reported that basil leaf contained saponins, flavonoids, steroids and tannins but had no alkaloids and terpenoids. The results of the present study showed that all metabolic secondary compounds contained in the selected medicinal herbs could be used to alter the hematological status and blood lipid profiles in broiler chickens.

The present study showed that *Sauropus androgynus* leaf contained 27.85 mg quercetin and 2.26 mg kaempferol/100 g, but no myricetin, apigenin and luteolin were detected. Andarwulan *et al.*[37] reported that *Sauropus androgynus* leaf contained 4.5 mg quercetin/100 g fresh weight, 138 mg kaempferol, <0.00002 mg myricetin, <0.006 mg leteolin and < 0.03 mg apigenin.

The present study showed that papaya leaves reduced WBC without altering other hematological variables. Thus, the present study contradicts the observation of Sheikh *et al.*[47], who reported that papaya leaf increased thrombocytes, MCH and MCV but had no effect on RBC, WBC, PCV and hemoglobin in rats. Moreover, papaya leaf extract has proven to be of significant importance in fighting coccidiosis, as shown in the reduction of oocyst counts and in improving RBC and WBC values in chickens[48]. It appears that the results of papaya leaf treatment differ according to the animal species treated.

A lymphocyte is one of the subtypes of WBC in a vertebrate's immune system. Thus, broiler chickens fed with *Sauropus androgynus* leaf may benefit by an increase in their immune system via an increase in lymphocytes without reducing WBC. The combination of *Elephantopus scaber* and *Sauropus androgynus* inclusion at certain doses increases the number of lymphocytes. Additionally, both have been found to be harmless to the developing fetus in pregnant mice[49]. It is interesting that broiler chickens fed a diet containing papaya leaf showed lower WBC but higher lymphocyte counts.

The number of RBCs in chickens ranges from 2.0×10^6 mm^{-3} to 3.5×10^6 mm^{-3} [48-50], hemoglobin ranges from 6.5-13.0 g mL^{-1} [50-52], PCV ranges from 22-43%[50,52,53], WBC ranges from 11.4-30 $\times10^3/mm3$[51,52]; lymphocyte counts range from 24-84%[52,53] and thrombocytes range from $3\text{-}33\times10^9$ L^{-1} [53], MCH 33.0-47.0 pg and MCHC 26.35[52]. Thus, the present study showed normal ranges of hematological status.

The present study agrees with the observation of Santoso *et al.*[29], who showed that the inclusion of medicinal herbs such as *Sauropus androgynus* had no effect on internal organ weight. Thus, medicinal herb inclusion did not have any side-effects on internal organ weight and did not increase toxicity in broiler chickens.

All medicinal herbs reduced plasma triglyceride in the present study. This biological activity might be due to the presence of phytoconstituents, i.e., flavonoids[54], tannins[55], saponins[56] and phenols[57]. Santoso *et al.*[3] reported that supplementation of unfermented *Sauropus androgynus* leaf extract reduced total cholesterol, triglyceride and LDL but increased HDL concentration in serum of layer chickens. Adenowo *et al.*[58] reported that papaya leaf supplementation reduced blood triglycerides in diabetic rats. Aljamal[59] reported that bay leaf inclusion reduced blood triglycerides in patients with type 1 diabetes. Husain *et al.*[60] reported that basil leaf extract reduced blood triglycerides in diabetic rats. Wang *et al.*[61] reported that noni juice reduced serum triglycerides in cigarette smokers. Halaby *et al.*[6] reported that *Moringa* inclusion reduced serum triglycerides in hyperlipidemic rats.

A lower LDL/HDL ratio in broiler chickens fed medicinal herb powder might be a beneficial indicator in reducing the risk of atherosclerosis. In addition, a high content of antioxidant and anti-inflammatory activities in medicinal herbs might inhibit LDL oxidation and therefore, they might inhibit atherosclerotic lesion development. Santoso *et al.*[3] reported that *Sauropus androgynus* leaf extract reduced atherogenic index, which indicated a lower risk of atherosclerosis.

Lipid fraction concentrations of broiler chickens were varied. Triglyceride, cholesterol, HDL and LDL levels ranged from 19.09-<150, 52-157.8 , >22-118.15 and 35.56-<130 mg dL^{-1}, respectively[62-65]. Therefore, the present study showed that lipid fraction concentrations in the plasma of broiler chickens were still within normal ranges.

This research might help to solve the problem of atherosclerosis either in animals or humans by using medicinal herbs either on their own or in combination with other medicinal herbs. In addition, the results of the present study might help to maintain broiler chickens without the need for antibiotics as a feed supplement and to develop organic farming for broiler chickens.

CONCLUSION

Sauropus androgynus leaf contains the highest protein content, at 34.37%, while noni fruit and basil leaf were rich in iron, at 7.80 and 6.43 ppm, respectively, followed by papaya leaf and *Sauropus androgynus* leaf, at 5.61 and 4.02 ppm, respectively. Papaya leaf reduced WBC but increased lymphocyte counts. All medicinal herbs showed lower LDL/HDL ratios and lower plasma triglyceride levels but higher HDL.

SIGNIFICANCE STATEMENT

This study discovers the possible uses of selected medicinal herbs as beneficial for reducing triglyceride, LDL and LDL/HDL ratios, which indicate a lower risk of atherosclerosis occurrence in broiler chickens. This study helps to reveal the critical area of atherosclerosis problems and organic farming that many researchers were unable to previously explore. Thus, a new theory may be created based on the usefulness of selected medicinal herbs and their combination on lowering the risk of atherosclerosis occurrence and developing organic farming for broiler chickens.

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