ISSN 1819-1878

Asian Journal of **Animal** Sciences



http://knowledgiascientific.com

Asian Journal of Animal Sciences

ISSN 1819-1878 DOI: 10.3923/ajas.2017.202.213



Research Article Efficacy of *Allium sativum* as Growth Promoter, Immune Booster and Cholesterol-lowering Agent on Broiler Chickens

¹Funmilayo Grace Adebiyi, ²Anthony Durojaiye Ologhobo and ³Isaac Oluseun Adejumo

¹College of Medicine, University of Ibadan, Ibadan, Nigeria

²Animal Biochemistry and Nutrition Research Unit, Department of Animal Science, University of Ibadan, Ibadan, Nigeria ³Laboratory of Animal Nutrition, Biotechnology and Food Safety, Department of Animal Science, Federal University Gashua, P.M.B. 1005, Gashua, Yobe State, Nigeria

Abstract

Background and Objective: Cholesterol plays important roles in membrane structure and as a precursor for the synthesis of the steroid hormones. However, abnormal deposition of cholesterol and cholesterol-rich lipoproteins in the coronary arteries could lead to atherosclerosis. This study was carried out to assess the efficacy of raw and sun-dried garlic on growth performance, immunity and cholesterol contents of broiler chickens. Materials and Methods: The efficacy of raw and sun-dried garlic as growth promoter, immune booster and cholesterol-lowering agent on broiler chickens was investigated in a 56 days feeding trial. Locally produced garlic bulbs were purchased from a commercial market in Northern Nigeria and ground with the husk into a paste. A portion of the paste was thinly spread on a clean mat and sun-dried for 2 days. The dried substance was ground to obtain dried garlic powder. The garlic paste and garlic powder were incorporated into broiler starter and finisher diets at 0, 1, 2 and 3% levels. The raw garlic was also included at 1, 2 and 3% levels (the inclusion level was calculated to exclude moisture content). Two hundred and ninety-four Anak 1 day old broiler chicks were randomly distributed into 7 dietary treatments of 21 birds each in a completely randomized design. Data were subjected to two-way analysis of variance using SPSS and the means were analysed using Duncan's multiple range test at statistical significance was p < 0.05. The white and red blood cells counts were determined using haemacytometer method. The packed cell volume and haemoglobin were measured by the methods of Wintrobe-micro-haematocrit and cyanomethaemoglobin, respectively. The serum samples were analyzed for total cholesterol, triglycerides and high-density lipoprotein cholesterol using Randox kit. Results: Raw and sun-dried garlic at 1, 2 and 3% did not influence growth performance and haematological parameters of broiler chickens but reduced the total cholesterol, liver cholesterol, triglycerides, abdominal fat pad and low-density lipoprotein cholesterol contents of the animals. Conclusion: The raw and sun-dried garlic at the inclusion levels reduced total cholesterol, liver cholesterol, triglycerides, abdominal fat pad and low-density lipoprotein cholesterol contents in the experimental animals while they improved high-density lipoprotein cholesterol.

Key words: Allium sativum, atherosclerosis, cholesterol, immune-booster, lipoprotein

Citation: Funmilayo Grace Adebiyi, Anthony Durojaiye Ologhobo and Isaac Oluseun Adejumo, 2017. Efficacy of *Allium sativum* as growth promoter, immune booster and cholesterol-lowering agent on broiler chickens. Asian J. Anim. Sci., 11: 202-213.

Corresponding Author: Isaac Oluseun Adejumo, Laboratory of Animal Nutrition, Biotechnology and Food Safety, Department of Animal Science, Federal University Gashua, P.M.B. 1005, Gashua, Yobe State, Nigeria Tel: +2348066446246

Copyright: © 2017 Funmilayo Grace Adebiyi *et al.* This is an open access article distributed under the terms of the creative commons attribution License, which permits unrestricted use, distribution and reproduction in any medium, provided the original author and source are credited.

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

Cholesterol is an important biological molecule that has roles in membrane structure as well as being a precursor for the synthesis of the steroid hormones. However, abnormal deposition of cholesterol and cholesterol-rich lipoproteins in the coronary arteries could lead to atherosclerosis, which is a major contributory factor in cardiovascular diseases. Hence, studies tailored towards reduction of cholesterol intake or its abnormal deposition in the coronary arteries is usually of interest. A recent increase in the popularity of alternative medicine and natural products has necessitated renewed interest in garlic and their derivatives as potential natural remedies. Garlic has been previously used as a cure for heart disease, headache and cancer, to improve the sexual condition as well as to cure snake bites as the norm of natural medicines is being often tried for many conditions. Garlic has been observed to contain about 2000 biologically active substances among which are volatile, water-soluble and oil-soluble organosulfur compounds, essential oils, dietary fiber, sugars 32% (included inulin), flavonoids and pectin¹⁻³.

Alliin is released when the garlic bulb is processed, crushed or minced and the released alliin interacts with alliinase. The product of hydrolysis and immediate condensation of the reactive intermediate (allylsulfenic acid) forms allicin (diallyl thiosulfinate) has been described to be stronger than some antibiotics⁴. It has been reported that essential oils from fresh bulbs of possess antimicrobial activity against *Staphylococcus aureus, Pseudomonas aeruginosa* and *Escherichia coli*⁵.

The use of garlic and its preparations includes being used as agents for prevention and treatment of cardiovascular diseases. Consumption of garlic could significantly lower blood pressure, prevent atherosclerosis, reduce serum cholesterol and triglyceride, inhibit platelet aggregation, as well as increase fibrinolytic activity⁶. Dietary inclusion of garlic powder has been reported to protect rats against gentamycin-induced hepatotoxicity, improve antioxidant status and modulate oxidative stress⁷. Extracts from garlic reduce lipid peroxidation and enhance antioxidant defense system⁸.

However, a lot of variations exist among garlic products which may be due to the amount of active ingredients influenced by method of preparation⁴. The review by Zeng *et al.*⁹ revealed that garlic is rich in compounds that have anticancer, antiinflammation, antiobesity, antidiabetes, antioxidants, antimicrobial activity, neuroprotective and immunological effects. Also, Li *et al.*¹⁰ observed that fresh garlic polysaccharide exhibited immunomodulatory activities. Limited information exists on the use of differently processed garlic as immune booster and growth promoter in chickens. It is believed that garlic is safe for consumption, relatively long-term trials may provide insights into the possible side-effects of different garlic and its products. Hence, this study was therefore designed to investigate the efficacy of raw and sun-dried garlic as growth promoter, immune boosters and cholesterol-lowering agent on broiler chickens.

MATERIALS AND METHODS

Source and preparation of garlic: The study was conducted between February and May, 2015. Locally produced garlic bulbs were purchased from a commercial market in Northern Nigeria and ground with the husk into a paste. A portion of the paste was thinly spread on a clean mat and sun-dried for 2 days. The dried substance was ground to obtain dried garlic powder. The garlic paste and garlic powder were incorporated into broiler starter and finisher diets at 0, 1, 2 and 3% levels. The raw garlic was also included at 1, 2 and 3% levels (the inclusion level was calculated to exclude moisture content). The compositions of the starter and finisher diets were presented in Table 1 and 2, respectively. All the diets were adjusted to be approximately isonitrogenous and isocaloric. The duration of the starter phase was 4 weeks (i.e., 1 day 4 weeks) while the finisher phase was between 4th and 8th week of the experimental period.

Feeding and management of birds: The experimental protocol and feeding trial was carried out in accordance with the principles of Laboratory Animal Care (NIH publication No.85-23, revised 1985) and as approved by the Animal Ethics Committee, Department of Animal Science, University of Ibadan, Nigeria. Two hundred and ninety-four Anak 1 day old broiler chicks obtained from a reputable hatchery in South-Western Nigeria were randomly distributed into 7 dietary treatments of 21 birds each in a completely randomized design. Each dietary treatment consisted of 6 replicates of 7 birds each. The chicks were reared in deep litter with feed and water supplied *ad libitum.* The management of the birds was as outlined by Oluyemi and Roberts¹¹. The feeding trial lasted for 56 days.

Experimental parameters measured: The weekly feed intake and weight gain were recorded from which feed conversion ratio was calculated during the 56 days experimental period. At the end of the experiment, 4 birds per replicate were sacrificed and the abdominal fat was weighed. The liver was also exercised and analyzed for total cholesterol:

Feed conversion ratio = Feed intake/Weight gain¹²

Asian J. Anim. Sci., 11 (5): 202-213, 2017

Table 1: Composition of the broilers starter diets (%)

	Diets						
Ingredients	1	2	3	4	5	6	7
Maize	51.00	52.00	52.50	53.00	52.00	52.50	53.00
Palm kernel cake	8.00	6.00	5.00	3.50	6.00	5.00	3.50
Wheat bran	4.00	3.00	2.00	1.50	3.00	2.00	1.50
Groundnut cake	10.00	11.00	11.00	11.50	11.00	11.00	11.50
Soya bean cake	20.50	20.50	21.00	21.00	20.50	21.00	21.00
Sun-dried garlic	0.00	1.00	9.00	3.00	0.00	0.00	0.00
Raw garlic	0.00	0.00	0.00	0.00	1.00	3.00	3.00
Fish meal	3.00	3.00	3.00	3.00	3.00	3.00	3.00
Bone meal	1.50	1.50	1.50	1.50	1.50	1.50	1.50
Oyster shell	1.20	1.20	1.20	1.20	1.20	1.20	1.20
Salt	0.25	0.25	0.25	0.25	0.25	0.25	0.25
Premix	0.25	0.25	0.25	0.25	0.25	0.25	0.25
Methionine	0.15	0.15	0.15	0.15	0.15	0.15	0.15
Lysine	0.15	0.15	0.15	0.15	0.15.	0.15	0.15
Calculated analyses							
Metabolizable energy (kcal kg ⁻¹)	2903.44	2901.98	2892.20	2880.60	2901.98	2892.20	2880.60
Crude protein (%)	22.49	22.51	22.42	22.34	22.51	79.47	22.34

Table 2: Composition of the broilers finisher's diets (%)

Diate

	Diets						
Ingredients	1	2	3	4	5	6	7
Maize	49.50	50.00	51.00	51.00	50.00	51.00	51.00
Palm kernel cake	7.50	7.00	7.50	6.00	7.00	7.50	6.00
Wheat bran	14.00	13.00	10.00	10.00	13.00	10.00	10.00
Groundnut cake	9.00	9.00	9.50	10.00	9.00	9.50	10.00
Soya bean cake	13.50	13.50	13.50	13.50	13.50	13.50	13.50
Sun-dried garlic	0.00	1.00	2.00	3.00	0.00	0.00	0.00
Raw garlic	0.00	0.00	0.00	0.00		2.00	3.00
Fish meal	3.00	3.00	3.00	3.00	3.00	3.00	3.00
Bone meal	1.50	1.50	1.50	1.50	1.50	1.50	1.50
Oyster shell	1.20	1.20	1.20	1.20	1.20	1.20	1.20
Salt	0.25	0.25	0.25	0.25	0.25	0.25	0.25
Premix	0.25	0.25	0.25	0.25	0.25	0.25	0.25
Methionine	0.15	0.15	0.15	0.15	0.15	0.15	0.15
Lysine	0.15	0.15	0.15	0.15	0.15	0.15	0.15
Calculated analyses							
Metabolizable energy (kcal kg ⁻¹)	2812.70	2800.30	2802.60	2783.10	2800.30	2802.6	2783.10
Crude protein (%)	20.35	20.14	20.05	20.00	20.14	20.05	20.00

Blood collection: Blood (5 mL) was collected through the jugular vein using needles and syringes from 3 birds per replicate on 21, 28, 42 and 56 days of the study. Blood (2 mL) for analysis of haematological parameters was collected in Ethylene Diamine Tetra acetic Acid (EDTA) bottles while the blood (3 mL) for analysis of serum lipids was collected into sterile sample tubes without anticoagulant. The serum was separated by centrifugation at 2000 rpm for 10 min. The sera were stored at -20°C until further analysis. The white and red blood cells counts were determined using haemacytometer method¹². The packed cell volume (PCV) and haemoglobin (Hb) were

measured by the methods of Wintrobe-micro-haematocrit and cyanomethaemoglobin, respectively¹³.

Analysis of serum lipids: The serum samples were analyzed for total cholesterol (TC), triglycerides (Tg) and high-density lipoprotein (HDL) cholesterol using Randox kit (CH20I, TR2IO and CH20₃) (Randox Laboratories Limited, Antrim, UK), respectively. The low-density lipoprotein (LDL) cholesterol was calculated using Friedewald formula¹⁴.

$$LDLc = TC - \left(HDLc - \frac{TG}{5}\right)$$

Estimation of liver cholesterol: At the end of the experimental period, 3 birds per replicate were randomly chosen for tissue collection. The birds were slaughtered, dressed and liver was carefully removed. The weight of the liver was taken and stored at -20°C until further analyses. Samples of the liver (1 g) per sample was homogenised manually in methanol-chloroform ($C_2H_5Cl_3$) at 2:1, v/v mixture using a Porter-Elvehgemhomogeniser (No. 7727, USA). The homogenate was made upto 20 fold (weight/volume) of the weight of the tissue samples. The clear extracts were assayed for total cholesterol, in well-labelled test tubes¹⁵. The Cholesterol Ouantitation Kit from Sigma (MAK043-IKT.Sigma Diagnostics, St. Louis, MO, USA) was used to assay the total cholesterol.

Carcass evaluation: The evaluation of carcass was done at the end of 56 days. Three birds per replicates were starved over-night and weighed before slaughtered. The starvation enhances better keeping quality, improved flavour of the meat as well as ridding the birds off feed present in the digestive track which may serve as contaminant during processing. The jugular vein was severed and bled before scalding in warm water for 1 min in order to improve the appearance and whiteness of the flesh. After scalding the feathers were removed by hand and the weight was taken. The carcass was eviscerated with a sharp knife. All intestinal organs and abdominal fat were carefully removed and Eviscerated weight was taken. The weights of liver and abdominal fat were recorded. Abdominal fat was expressed as percentage of the eviscerated weights. **Chemicals analyses:** The test ingredients i.e., the raw garlic and sun-dried garlic and the test diets were subjected to chemical analyses for the determination of nitrogen, ether extract, crude fibre, ash and moisture by the procedures of the Association of Official Analytical Chemist¹⁶.

Statistical analysis: Data were subjected to two-way analysis of variance (ANOVA) using SPSS (Version 21.0. Armonk, NY: IBM Corp.) and the means were separated using Duncan's multiple range test (DMRT) of the same software. Values are expressed as mean (standard error of mean (SEM)). The level of statistical significance was p<0.05.

RESULTS

The proximate compositions of raw and dried garlic are stated thus, crude protein: 5.74% (raw), 8.36% (dried), ether extract: 1.02% (raw), 1.28% (dried), fiber: 0.76% (raw), 0.63% (dried), ash: 2.18% (raw), 3.28% (dried), dry matter: 36.8% (raw), 79.8% (dried) and gross energy: 1.42 Cal g^{-1} (raw), 1.29 Cal g⁻¹ (dried). The proximate analysis of the broiler starter and finisher diets are presented in Table 3 and 4, respectively. The nutritional values of raw garlic (RG) per 100 g of sample were lower than the sun-dried garlic (SDG) powder. The value of the crude protein of SDG powder was 31.34% higher than RG. The values of ether extract, ash and dry matter were also higher in SDG powder, while the experimental diets for all the treatments were iso-nitrogenious and iso-caloric. Average feed intake, average weight gain and feed conversion ratio were not significantly different across the treatments.

Diets	Crude protein (%)	Ether extract (%)	Crude fiber (%)	Ash (%)	Dry matter (%)	Gross energy (Cal g ⁻¹)
1 (Control)	22.70	3.92	3.68	8.62	89.63	3.012
2 (SDG 1%)	22.04	4.01	3.74	9.81	89.74	3.036
3 (SDG 2%)	22.83	3.84	3.65	9.74	89.82	3.027
4 (SDG 3%)	22.46	4.16	3.79	9.92	89.77	3.038
5 (RG 1%)	22.97	3.97	3.83	9.79	89.69	3.074
6 (RG 2%)	22.35	4.21	3.76	9.85	89.71	3.096
7 (RG 3%)	22.56	4.32	3.81	9.97	89.65	3.082

SDG: Sun-dried garlic, RG: Raw garlic

Table 4: Proximate analysis of experimental diets (finisher phase)

Crude protein (%)	Ether extract (%)	Crude fiber (%)	Ash (%)	Dry matter (%)	Gross energy (Cal g ⁻¹)
19.68	3.56	3.77	9.04	89.65	3.016
19.84	3.62	4.14	9.16	89.69	3.019
19.96	3.48	4.23	9.24	89.87	3.024
19.72	3.65	4.18	9.09	89.26	3.015
19.63	3.74	4.25	9.96	89.49	3.032
19.54	3.53	4.29	9.27	89.56	3.028
19.48	3.59	4.16	9.29	89.64	3.022
	19.68 19.84 19.96 19.72 19.63 19.54	19.68 3.56 19.84 3.62 19.96 3.48 19.72 3.65 19.63 3.74 19.54 3.53 19.48 3.59	19.68 3.56 3.77 19.84 3.62 4.14 19.96 3.48 4.23 19.72 3.65 4.18 19.63 3.74 4.25 19.54 3.53 4.29 19.48 3.59 4.16	19.683.563.779.0419.843.624.149.1619.963.484.239.2419.723.654.189.0919.633.744.259.9619.543.534.299.2719.483.594.169.29	19.68 3.56 3.77 9.04 89.65 19.84 3.62 4.14 9.16 89.69 19.96 3.48 4.23 9.24 89.87 19.72 3.65 4.18 9.09 89.26 19.63 3.74 4.25 9.96 89.49 19.54 3.53 4.29 9.27 89.56 19.48 3.59 4.16 9.29 89.64

SDG: Sun-dried garlic, RG: Raw garlic

Figure 1 shows the weekly representation of feed intake and blood parameters of chickens fed with 0, 1, 2 and 3% sun-dried garlic and raw garlic over a period of 56 days while Table 5 shows the influence of dietary garlic on feed intake, body weight gain and feed conversion ratio on birds fed SDG and RG over a period of 56 days. Dietary inclusion of garlic for both RG and SDG did not affect feed intake, weight gain and feed conversion ratio significantly. The effect of intake of both RG and SDG powder by broilers did not significantly affect the packed cell volume, hemoglobin, red blood cell counts, white blood cell counts, neutrophils and lymphocytes.

The effect of feeding 1, 2 and 3% RG and SDG to broilers on total cholesterol is shown in Fig. 2. Birds fed garlic had consistently lower values of total cholesterol compared with birds fed control diet as measured over the 8 weeks experimental period. The highest value of 122 mg dL⁻¹ was recorded at week 3 for birds fed control diet while bird fed 2% RG had the lowest value (89.9 mg dL⁻¹) recorded at week 3. The experimental duration did not affect the TC.

Birds fed 2% RG had significantly (p<0.05) lower mean value of 96.1 mg dL⁻¹ compared with birds fed other diets. While birds fed with the control diet recorded the highest value of TC as averaged over the 8 weeks experimental period. Generally, birds fed either RG or SDG had a significantly (p<0.05) lower value of TC compare to the control diet (Table 5).

The effect of dietary treatments on triglyceride is shown in Fig. 2 and Table 5. There was a general decrease in the values of Tg of all the treatment groups as the experiment progressed. However, there was a slight increase in the 8th week for birds fed 2 and 3% SDG and 2 and 3% RG. Birds fed control diet and 1% SDG recorded a steady decrease up to the 8th week. The highest value of Tg (9I.4 mg dL⁻¹) was recorded for birds fed the control diet at week 4 while lowest value of 35.2 mg dL⁻¹ recorded for birds fed 2% SDG at week 6. On the average, different levels of garlic had a significant effect on the values Tg of the birds. Birds fed 2% SDG had a significantly (p<0.05) lower value of 53.3 mg dL⁻¹ compared to birds fed other diets as averaged over 8 weeks (Table 5). There was however, no significant difference in values of Tg of birds fed 1% RG (63.3 mg dL⁻¹) and 1% SDG (60.8 mg dL⁻¹). These values were however, significantly (p<0.05) higher than the Tg of birds fed diets 3, 4, 6 and 7. The Tg value of birds fed 3% SDG and 2 and 3% RG were not significantly different as averaged over 8 weeks (Table 5).

Figure 2 shows the weekly representation of lymphocytes and lipid parameters of chickens fed with 0, 1, 2 and 3% sun-dried garlic and raw garlic over a period of 56 days. The values of HDL for all the treatment groups increased with age

Table 5: Effect of dietary inclusion of varying levels of raw and su	levels of raw and sun-drie	ed garlic on the lipid para	in-dried garlic on the lipid parameters of broiler chickens	S			
	Treatments						
Parameters	1 (Control)	2 (1% SDG)	3 (2% SDG)	4 (3% SDG)	5 (1% RG)	6 (2% RG)	7 (3% RG)
Triglycerides (mg dL ⁻¹)	68.70 (19.96) ^a	63.30 (15.20) ^b	53.30 (18.85) ^d	56.80 (18.38) ^c	60.80 (20.02) ^b	56.80 (18.15) ^c	57.60 (18.21) ^c
Cholesterol (mg dL ⁻¹)	$116.50 (6.69)^{a}$	108.40 (8.87) ^{bc}	105.40 (9.61) ^c	106.40 (8.43) ^{bc}	110.80 (6.28) ^b	96.10 (7.76) ^d	106.40 (10.5I) ^{bc}
HDL-cholesterol (mg dL ⁻¹)	$25.30 (4.97)^{a}$	31.50 (5.37) ^b	36.90 (4.91) ^{de}	35.10 (6.08) ^{cd}	32.10 (5.86) ^b	37.70 (5.95)c	34.80 (5.84)⁵
LDL-cholesterol (mg dL ⁻¹)	77.50 (8.21) ^a	64.20 (9.17) ^{bc}	57.90 (11.10) ^d	59.90 (9.25) ^{cd}	66.90 (7.43) ^b	47.00 (6.92) ^c	60.10 (11.65) ^{cd}
Abdominal fat pad (%)	1.76 (0.039) ^a	∞0.67 (0.099)	0.53 (0.058) ^{cd}	0.31 (0.017) ^e	0.76 (0.069) ^b	0.45 (0.063) ^{de}	0.35 (0.038) ^{de}
Liver cholesterol (mg/100 g wet tissue)	296.40 (7.95) ^a	229.90 (10.40) ^b	207.10 (3.81) ^b	138.10(7.29) ^d	173.50 (7.31)⁵	139.80 (7.58) ^d	121.40 (8.47) ^d
Values are means (standard deviation), means with the same sup	is with the same superscr	ipts within the same row	berscripts within the same row are not significantly different at p<0.05	rent at p<0.05			

SDG: Sun-dried garlic, RG: Raw garlic

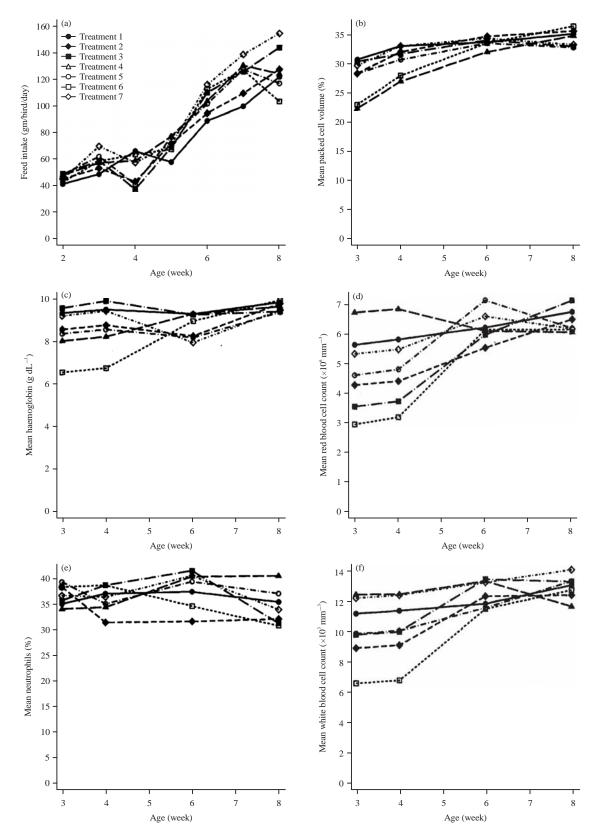


Fig. 1(a-f): Weekly representation of feed intake and blood parameters of chickens fed with 0, 1, 2 and 3% sun-dried garlic and raw garlic over a period of 56 days, (a) Mean feed intake, (b) Mean packed cell volume, (c) Mean haemoglobin levels, (d) Mean red blood cell counts, (e) Mean neutrophils levels and (f) Mean white blood cell counts

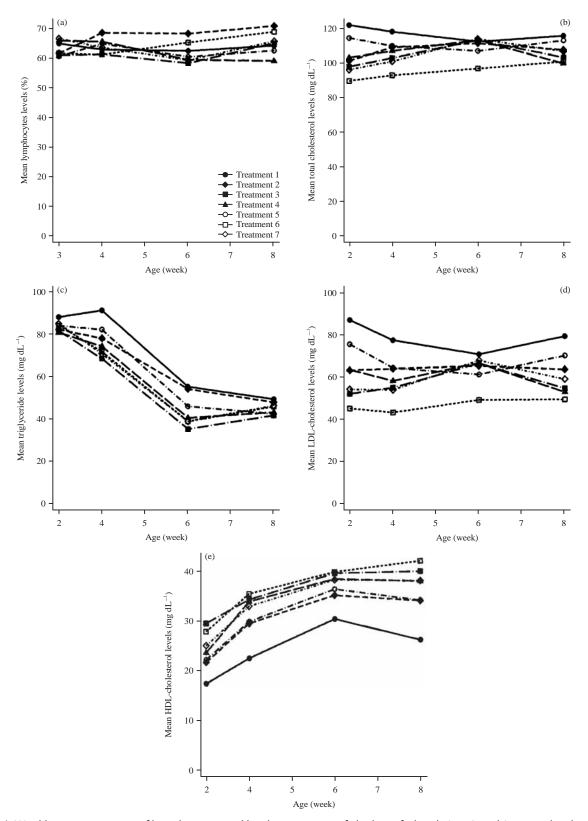


Fig. 2(a-e): Weekly representation of lymphocytes and lipid parameters of chickens fed with 0, 1, 2 and 3% sun-dried garlic and raw garlic over a period of 56 days, (a) Mean lymphocyte levels by age, (b) Mean cholesterol levels by age, (c) Mean triglyceride levels by age, (d) Mean low-density lipoprotein cholesterol levels by age and (e) Mean high-density lipoprotein cholesterol levels by age

up to the 6th week except for birds fed 2% SDG and 2% RG which showed steady increase throughout the experimental period. The highest value of 42.3 mg dL⁻¹ was recorded for birds fed 2% RG at week 8. However, birds fed control diets recorded the lowest value of HDL (17.5 mg dL⁻¹) at week 2, birds fed control diets consistently recorded lower values when compared with birds fed other diets throughout the experimental period. Table 5 shows the mean values of HDL of birds fed graded levels of garlic. Birds fed control diets had a significantly (p<0.05) lower mean value of 25.3 mg dL⁻¹ compared with bird fed different levels of RG and SDG.

The values of HDL recorded for birds fed with 2 and 3% SDG and RG when compared with birds fed with the control, 1% RG and 1% SDG diets were significantly (p<0.05) higher. The highest value of HDL was recorded for birds fed with 2% RG (37.7 mg dL⁻¹).

The LDL values for all the treatment groups were not consistent in any particular trend. The observable trend was that the LDL tend to decrease as the experiment progressed. The mean LDL for birds fed with the control diet was significantly (p<0.05) higher than birds fed with graded levels of RG and SDG (Table 5). The reducing effect of garlic on LDL was more pronounced when 2% of RG was included in the diets. However, a reduction of 17.2, 25.3 and 22.7% was observed in birds fed with 1, 2 and 3% SDG. Inclusion of 1, 2 and 3% of RG in the diets also reduced the LDL values by 13.7, 39 and 22.5%, respectively (Table 5).

The AFP of broilers fed with various inclusion of RG and SDG is shown in Table 5. The effect of RG and SDG on AFP was such that birds fed with the control diet recorded the highest value of AFP. The values of AFP of birds fed with 1% RG and SDG were similar but significantly higher (p<0.05) than the AFP of birds fed with 3% SDG, 2% RG and 3% RG. The lowest value of AFP was observed in birds fed with 3% SDG. The effect of the different levels of RG and SDG on liver cholesterol is shown in Table 5. The mean liver cholesterol decreased with an increase in both RG and SDG. The lowest mean liver cholesterol (121.4 mg/100 g tissue) was recorded by the birds fed with 3% RG. This value was significantly lower than the LC of birds fed with the control diet, 1 and 2% RG and SDG.

DISCUSSION

The non-significance of mean feed intake, weight gain and feed conversion ratio of broiler chickens may imply that inclusion of either RG or SDG in broiler diets up to 3% are not likely to have adverse effect on the growth performance of broiler chickens. Hence, the active ingredients in both raw and sun-dried garlic could be said not to enhance the palatability of the experimental birds. These results agree with the findings of Rahmatnejad et al.¹⁷, who reported that garlic given at 1000 g t⁻¹ did not affect feed intake, weight gain and feed conversion ratio in broiler chicken. The findings of similar studies conducted by Onibi et al.18 and Fadlalla et al.19 reported no significant differences in weight gain of broiler chickens fed with 500 and 5000 mg kg⁻¹ garlic powder and 0.15, 0.3, 0.45 and 0.6% garlic powder, respectively. However, studies conducted by some researchers differ somewhat from the results observed in this study. Such results included those of Rahardja et al.20, who reported that inclusion of 1.2 and 4.0% levels of garlic in feed significantly increased feed intake of laying hens in a 4 weeks trial. Likewise, Kumar et al.21 reported that garlic active based growth promoter given at 250 ppm, significantly increased the body weight of broiler chickens by day 42 of the trial. Similarly, increased feed intake, weight gain and feed conversion efficiency with garlic supplementation at 250 mg kg⁻¹ body weight have been reported in a 2 month trial in new born calves up to their 2 month pre-ruminant stage²². In contrast to the above reports, Nidaullah et al.23 reported a reduction in the feed intake of broiler birds fed garlic supplement. Similarly, the inclusion of 0.1 and 0.2% garlic and thyme in layers' diets significantly decreased the feed intake by the 6th week²⁴. Cullen et al.²⁵ also reported a reduction in feed intake in pigs fed 1 and 10 g kg⁻¹ garlic diet during the grower phase.

The inconsistency in the results of various studies¹⁷⁻²⁵ on the effect of various forms of garlic on chickens' productive performance might be due to the use of different levels of garlic in the diets which could affect the available active ingredients in the diets of experimental birds. In addition, different forms of garlic preparation may also result in different active ingredients of the processed garlic. Such active component of garlic could therefore exact different effect on the test animals. The feed conversion ratio was not affected by inclusion of dietary RG and SDG in broilers' diets. Therefore, inclusion of both RG and SDG in the diet of broilers suggests that active ingredients in garlic do not have negative effect on the feed conversion ratio of broiler chicken. This observed values of FCR in the present study agrees with the findings of Aji et al.26, who recorded no significant difference in FCR of day old broiler chicks orally given 25, 50 and 100 mg of dissolved garlic powder for 21 days. In another study conducted by Khan et al.27, who reported no significant difference in feed consumption and feed efficiency despite significant increase in weight gain of laying hens whose diets were supplemented with 2, 4, 6 and 8% oven dried garlic powder.

The values of haematological parameters obtained in this study were not significantly affected by inclusion of varying levels of RG and SDG in the broiler diets. However, these values are within the normal range for adult chicken as recorded by Mitruka and Rawnsley²⁸, who reported 26-45.2% PCV for adult chicken. Although there was a slight drop of PCV in birds fed 3% SDG and 3% RG at week 3 when compared with the values obtained by Mitruka and Rawnsley²⁸. The slight differences in the value of PCV might due to the genetic make-up of birds. However, these values fall within the range value of PCV recorded by Bamidele and Adejumo²⁹, who observed a range of 18.00-27.50% in pullet fed a mixture of garlic and ginger powder. The concentration of Hb obtained ranged between 6.5 and 9.9 g dL $^{-1}$ which could be considered normal when compared with the normal range of Hb of adult chicken (7.50 and 13.10 g dL⁻¹) reported by Mitruka and Rawnsley²⁸. The combination of duration and treatments effects had no significant effect on the broiler chickens. It could therefore be observed that the amount of both SDG and RG used in this experiment did not exert adverse effect on the Hb concentrations of broiler chickens. The results of Hg agree with the work of Ademola et al.30, who recorded insignificant differences in HB of broilers fed 1, 1.5 and 2% garlic powder.

The red blood cell counts were not significantly affected by varying levels of both SDG and RG inclusion in the broilers' diets. Although birds fed control diet, 1 and 2% SDG and 2% RG recorded increasing values in RBC with age. These results corroborate the reports of Islam *et al.*³¹, who observed that Hb of chickens increased with age. However, the slight decrease in the values of RBC of birds fed 3% SDG, 1 and 3% RG at week 8 was not significant. Oboh³² also reported that inclusion of 4% garlic in feed could cause haemolytic anaemia. It is therefore necessary to note that inclusion of high level of garlic could cause adverse effect to the concentrations of PCV, Hb and RBC as reports showed that ingestion of garlic causes anaemia³³.

The white blood cell (WBC), Lymphocyte and Neutrophils levels were not significantly affected by SDG and RG. This indicates that the treatment did not influence immune response of the broiler chickens. This study agreed with Pourali *et al.*³⁴, who reported a non-significant effect of supplementation of 0.2, 0.4, 0.6, 0.8 and 1% garlic powder on newcastle antibody titer of broiler chicken over a period of 42 days. These results also corroborate the findings of Jafari *et al.*³⁵ that inclusion of garlic powder to the diet of broilers had no beneficial effect on immune response.

In contrast with these results, Birrenkott *et al.*³⁶ reported that inclusion of garlic in laying hens' diet resulted in increased total WBC counts. In a related experiment, Fadlalla *et al.*¹⁹

reported that the inclusion of garlic powder caused increased in the total WBC counts of broiler chickens. This discrepancy may likely relate to the variety of garlic, preparation, process, dose, variety of garlic products, strains of experimental birds and duration of the experiment. Gorinstein *et al.*³⁷ noted that the contents of bioactive compounds in the herbs are influenced by geographical region, weather, storage conditions and their degree of ripeness.

Levels of TC, Tg and LDL decreased as the inclusion levels of both raw and sun-dried garlic increased in the diets. The highest reduction of TC was observed in birds fed 2% raw garlic. The RG and SDG significantly reduced the values of serum TC, Tg and LDL. A similar pattern was observed in the values of Tg and LDL in the study conducted by Chi *et al.*³⁸, who reported 30% reduction in plasma cholesterol when rats were fed with diets supplemented with 2 or 3% garlic powder while Chowdhury *et al.*³ recorded 15, 28, 33 and 43% reduction in serum cholesterol when laying hens were fed 2, 4, 6 and 8% garlic paste, respectively but no further reduction was observed when birds were fed 10% inclusion of garlic paste. However, the current study showed that inclusion of both 2% of raw garlic and sun-dried garlic powder in broiler chicks was enough to significantly reduce TC.

The results of this present study also corroborate the observations of Issa *et al.*³⁹, who recorded that TC, Tg and LDL were significantly decreased in broiler chicken fed with 0.2% and 0.4% garlic powder when compared with birds fed with control diet. Similar findings were also recorded by Bamidele and Adejumo²⁹, who found a significant reduction in the value of LDL when the diets of growing pullets were supplemented with a mixture of garlic and ginger powder.

The significant reductions observed in TC, Tg and LDL cholesterol concentrations in this study are probably due to the fact that garlic increases excretion of bile acids and depressed the hepatic activities of lipogenic and cholesterogenic enzymes such as malic enzyme, fatty acid synthase, glucose-6-phosphate de-hydrogenase and 3-hydroxy-3-methyl-glutary-CoA (HMG CoA) reductase⁴⁰. Also, Chi *et al.*³⁸ reported that garlic increased the excretion of acidic and neutral steroids in both 16 and 10 weeks old rats after feeding rats with 2 and 4% lyophilised garlic diets.

The results obtained in the present study also showed that the inclusion of RG and SDG significantly (p<0.05) increased the value of HDL cholesterol. The HDL cholesterol concentrations tended to follow an inverse pattern from other lipids. These observations in the serum lipids in this study demonstrate the ability of both raw and sun-dried garlic intake by broiler chickens to reduce the serum TC, Tg and LDL

cholesterol while favouring a rise in the serum HDL cholesterol concentrations. The findings in this study is similar to the observation of Choi *et al.*⁴¹, who reported a significant increase in the value of HDL when broiler chickens were fed with 3 and 5% garlic powder. Likewise, Ademola *et al.*³⁰ recorded an increase of 20.03, 15.79 and 23.77% in HDL cholesterol when broiler chickens were fed 1, 1.5 and 2% sun-dried garlic powder.

The results of the present study show that intake of RG and SDG significantly (p<0.05) reduced the abdominal fat pad (AFP) in broiler chickens. It was observed in this study that the weight of the AFP tended to concur with the values of serum Tg and liver cholesterol in response to inclusion of RG and SDG in broilers diet. The same trend was also observed by Shahriari et al.42, who recorded decrease in AFP when 2 and 4% garlic powder were included in broilers' diets. These researchers opined that the Tg secreted in the liver are the main source of plasma Tg, therefore, simultaneous reduction in the weight of the AFP with levels of serum and liver Tg may be due to the effect of garlic suppressing hepatic TG and total cholesterol synthesis. The reduction in the AFP weights recorded in this study also agrees with earlier studies conducted by Dieumou et al.43 and Ademola et al.30, who fed broiler chickens with fed 40 ppm and 60 ppm garlic organic extract and 1, 1.5 and 2% garlic powder, respectively and observed significant reduction in the values of AFP.

Inclusion of RG and SDG in diet of the broiler chickens reduced the level of liver cholesterol (LC). The mean LC decreased with increased level of both RG and SDG. This is in agreement with the findings of Konjufca *et al.*⁴⁴, who recorded significant reduction in LC when broilers were fed garlic powder. The reduction in LC observed in this study could be as a result of the ability of some component of garlic to inhibit synthesis of cholesterol in the liver. Garlic has been confirmed by Konjufca *et al.*⁴⁴ and Qureshi *et al.*⁴⁵ to reduce the activities of HMG-CoA reductase enzyme and cholesterol 70-hydroxylase by about 40% in the microsomes of birds fed garlic.

CONCLUSION AND FUTURE RECOMMENDATION

Inclusion of raw and sun-dried garlic at 1, 2 and 3% did not influence growth performance and haematological parameters of broiler chickens in this study. However, the raw and sun-dried garlic at the inclusion levels reduced total cholesterol, liver cholesterol, triglycerides, abdominal fat pad and low-density lipoprotein cholesterol contents in the experimental animals while they improved high-density lipoprotein cholesterol. Further studies may be necessary to establish the use of garlic as growth promoter as well as immune boosters in animal nutrition.

SIGNIFICANCE STATEMENTS

This study proofs the effect of differently processed garlic as growth enhancer and immune booster in broiler chickens. This study will help researchers to compare the hypo-cholesterolemic effect of differently processed garlic for use in human and animal nutrition.

REFERENCES

- 1. Cerny, C. and R. Guntz-Dubini, 2013. Formation of cysteine-S-conjugates in the Maillard reaction of cysteine and xylose. Food Chem., 141: 1078-1086.
- Swiderski, F., M. Dabrowska, A. Rusaczonek and B. Waszkiewicz-Robak, 2007. Bioactive substances of garlic and their role in dietoprophylaxis and dietotherapy. Rocz. Panstw. Zakl. Hig., 58: 41-46.
- 3. Chowdhury, S.R., S.D. Chowdhury and T.K. Smith, 2002. Effects of dietary garlic on cholesterol metabolism in laying hens. Poult. Sci., 81: 1856-1862.
- 4. Majewski, M., 2014. *Allium sativum*: Facts and myths regarding human health. Roczniki Panstwowego Zakladu Higieny, 65: 1-8.
- Casella, S., M. Leonardi, B. Melai, F. Fratini and L. Pistelli, 2013. The role of diallyl sulfides and dipropyl sulfides in the *in vitro* antimicrobial activity of the essential oil of garlic, *Allium sativum* L. and leek, *Allium porrum* L. Phytother. Res., 27: 380-383.
- 6. Chan, J.Y.Y., A.C.Y. Yuen, R.Y.K. Chan and S.W. Chan, 2013. A review of the cardiovascular benefits and antioxidant properties of allicin. Phytother. Res., 27: 637-646.
- Ademiluyi, A.O., G. Oboh, T.R. Owoloye and O.J. Agbebi, 2013. Modulatory effects of dietary inclusion of garlic (*Allium sativum*) on gentamycin-induced hepatotoxicity and oxidative stress in rats. Asian Pac. J. Trop. Biomed., 3: 470-475.
- El-Kott, A.F., A.M. Abdel-Aziz, A.E.K.M. Abd El-Latif, E.M. El-Gamal and A.M. Khalil, 2012. Amelioration of nitrate-induced hepatotoxicity by *Allium sativum* in mice. J. Med. Sci., 12: 85-91.
- 9. Zeng, Y., Y. Li, J. Yang, X. Pu and J. Du *et al.*, 2017. Therapeutic role of functional components in Alliums for preventive chronic disease in human being. Evidence-Based Complement. Altern. Med. 10.1155/2017/9402849.
- Li, M., Y.X. Yan, Q.T. Yu, Y. Deng and D.T. Wu *et al.*, 2017. Comparison of immunomodulatory effects of fresh garlic and black garlic polysaccharides on RAW 264.7 macrophages. J. Food Sci., 82: 765-771.

- 11. Oluyemi, J.A. and F.A. Roberts, 2000. Poultry Production in Warm Wet Climates. 2nd Edn., Spectrum Books, Ibadan, Nigeria, ISBN-13: 9789780290979, pp: 18-34.
- USAID., 2011. Feed Conversion Ratio (FCR): How to calculate it and how it is used? USAID Technical Bulletin No. 7, USAID-Harvest, Phnom Penh, Cambodia, December 2011, pp: 1-2.
- Ghai, C.L., 1993. Haematology. In: A Textbook of Practical Physiology, Ghai, C.L. (Ed.). Jaypee Brothers Medical Publishers (Pvt.) Ltd., New Delhi, India, pp: 119-202.
- 14. Friedewald, W.T., R.I. Levy and D.S. Fredrickson, 1972. Estimation of the concentration of low-density lipoprotein cholesterol in plasma, without use of the preparative ultracentrifuge. Clin. Chem., 18: 499-502.
- Searcy, R.L., L.M. Bergquist, R.C. Jung, R. Craig and J. Korotzer, 1960. Simplified technic for estimation of serum lipoprotein cholesterol. Clin. Chem., 6: 585-597.
- AOAC., 2005. Official Methods of Analysis. 18th Edn., Association of Official Analytical Chemists, Washington, DC., USA.
- Rahmatnejad, E., H. Roshanfekr, O. Ashayerizadeh, M. Mamooee and A. Ashayerizadeh, 2009. Evaluation the effect of several non-antibiotic additives on growth performance of broiler chickens. J. Anim. Vet. Adv., 8: 1757-1760.
- Onibi, G.E., O.E. Adebisi, A.N. Fajemisin and A.V. Adetunji, 2009. Response of broiler chickens in terms of performance and meat quality to garlic (*Allium sativum*) supplementation. Afr. J. Agric. Res., 4: 511-517.
- Fadlalla, I.M.T., B.H. Mohammed and A.O. Bakhiet, 2010. Effect of feeding garlic on the performance and immunity of broilers. Asian J. Poult. Sci., 4: 182-189.
- Rahardja, D.P., M.R. Hakim, W. Pakiding and V.S. Lestari, 2010. Hypocholesterolemic effect of garlic powder in laying hen: Low cholesterol egg? J. Indonesian Trop. Anim. Agric., 35: 16-21.
- Kumar, S., K.C. Sharadamma and P.M. Radhakrishna, 2010. Effects of a garlic active based growth promoter on growth performance and specific pathogenic intestinal microbial counts of broiler chicks. Int. J. Poult. Sci., 9: 244-246.
- 22. Ghosh, S., R.K. Mehla, S.K. Sirohi and B. Roy, 2010. The effect of dietary garlic supplementation on body weight gain, feed intake, feed conversion efficiency, faecal score, faecal coliform count and feeding cost in crossbred dairy calves. Trop. Anim. Health Prod., 42: 961-968.
- Nidaullah, H., F.R. Durrani, S. Ahmad, I.U. Jan and S. Gul, 2010. Aqueous extract from different medicinal plants as anticoccidial, growth promotive and immunostimulant in broilers. ARPN J. Agric. Biol. Sci., 5: 53-59.

- 24. Ghasemi, R., M. Zarei and M. Torki, 2010. Adding medicinal herbs including garlic (*Allium sativum*) and thyme (*Thymus vulgaris*) to diet of laying hens and evaluating productive performance and egg quality characteristics. Am. J. Anim. Vet. Sci., 5: 151-154.
- 25. Cullen, S.P., F.J. Monahan, J.J. Callan and J.V. O'Doherty, 2005. The effect of dietary garlic and rosemary on grower-finisher pig performance and sensory characteristics of pork. Irish J. Agric. Food Res., 44: 57-67.
- Aji, S.B., K. Ignatius, A.A.Y. Ado, J.B. Nuhu and A. Abdulkarim et al., 2011. Effects of feeding onion (*Allium cepa*) and garlic (*Allium sativum*) on some performance characteristics of broiler chickens. Res. J. Poult. Sci., 4: 22-27.
- 27. Khan, S.H., S. Hasan, R. Sarda and M.A. Anjum, 2008. Effects of dietary garlic powder on cholesterol concentration in native desi laying hens. Am. J. Food Technol., 3: 207-213.
- Mitruka, B.M. and H.M. Rawnsley, 1977. Clinical Biochemical and Hematological Reference Values in Normal Experimental Animals. Masson Publ., New York, USA., ISBN-13: 9780893520069, Pages: 272.
- 29. Bamidele, O. and I.O. Adejumo 2012. Effect of garlic (*Allium sativum* L.) and ginger (*Zingiber officinale* Roscoe) mixtures on performance characteristics and cholesterol profile of growing pullets. Int. J. Poult. Sci., 11: 217-220.
- Ademola, S.G., T.E. Lawal, O.O. Egbewande and G.O. Farinu, 2012. Influence of dietary mixtures of garlic and ginger on lipid composition in serum, yolk, performance of pullet growers and laying hens. Int. J. Poult. Sci., 11: 196-201.
- Islam, M.S., N.S. Lucky, M.R. Islam, A. Ahad, B.R. Das, M.M. Rahman and M.S.I. Siddiui, 2004. Haematological parameters of fayoumi, assil and local chickens reared in Sylhet region in Bangladesh. Int. J. Poult. Sci., 3: 144-147.
- 32. Oboh, G., 2004. Prevention of garlic-induced hemolytic anemia using some tropical green leafy vegetables. J. Med. Food., 7: 498-501.
- Nakagawa, S., K. Masamoto, H. Sumiyoshi, K. Kunihiro and T. Fuwa, 1980. Effect of raw and extracted-aged garlic juice on growth of young rats and their organs after peroral administration. J. Toxicol. Sci., 5: 91-112, (In Japanese).
- 34. Pourali, M., S.A. Mirghelenj and H. Kermanshahi, 2010. Effects of garlic powder on productive performance and immune response of broiler chickens challenged with Newcastle Disease Virus. Global Vet., 4: 616-621.
- Jafari, R.A., M. Razi-Jalali, M. Ghorbanpoor and S.M.R.M. Saraei, 2008. Effect of dietary garlic on immune response of broiler chicks to live Newcastle disease vaccine. Pak. J. Biol. Sci., 11: 1848-1851.
- Birrenkott, G., G.E. Brockenfelt, M. Owens and E. Halpin, 2000. Yolk and blood cholesterol levels and organoleptic assessment of eggs from hens fed a garlic-supplemented diet. Poult. Sci., 79(Suppl. 1): 75-75.

- 37. Gorinstein, S., H. Leontowicz, M. Leontowicz, Z. Jastrzebski and K. Najman *et al.*, 2010. The influence of raw and processed garlic and onions on plasma classical and non-classical atherosclerosis indices: Investigations *in vitro* and *in vivo*. Phytother. Res., 24: 706-714.
- Chi, M.S., E.T. Koh and T.J. Stewart, 1982. Effects of garlic on lipid metabolism in rats fed cholesterol or lard. J. Nutr., 112: 241-248.
- 39. Issa, K.J. and J.M. Abo Omar, 2012. Effect of garlic powder on performance and lipid profile of broilers. Open J. Anim. Sci., 2: 62-68.
- 40. Yeh, Y.Y. and L. Liu, 2001. Cholesterol-lowering effect of garlic extracts and organosulfur compounds: Human and animal studies. J. Nutr., 131: 989S-993S.
- Choi, I.H., W.Y. Park and Y.J. Kim, 2010. Effects of dietary garlic powder and α-tocopherol supplementation on performance, serum cholesterol levels and meat quality of chicken. Poult. Sci., 89: 1724-1731.

- 42. Shahriari, A., R.F. Tabatabaie, R.A. Jafari and B. Ghorbanzadeh, 2009. Modulation of serum and liver triglyceride and abdominal fat pad weight by dietary garlic in male broilers. Int. J. Vet. Res., 3: 101-105.
- Dieumou, F.E., A. Teguia, J.R. Kuiate, J.D. Tamokou, N.B. Fonge and M.C. Dongmo, 2009. Effects of ginger (*Zingiber officinale*) and garlic (*Allium sativum*) essential oils on growth performance and gut microbial population of broiler chickens. Livest. Res. Rural Dev., Vol. 21, No. 8.
- 44. Konjufca, V.H., G.M. Pesti and R.I. Bakalli, 1997. Modulation of cholesterol levels in broiler meat by dietary garlic and copper. Poult. Sci., 76: 1264-1271.
- 45. Qureshi, A.A., Z.Z. Din, N. Abuirmeileh, W.C. Burger, Y. Ahmad and C.E. Elson, 1983. Suppression of avian hepatic lipid metabolism by solvent extracts of garlic: Impact on serum lipids. J. Nutr., 113: 1746-1755.