Lipid Profile of Chicken (*Gallus domesticus*) in Response to Dietary Supplementation of Garlic (*Allium sativum*)

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**Abstract:** Garlic is widely distributed and used in all parts of the world as a spice and herbal remedy for various ailments, including its role in diabetes, blood coagulation, metabolism and immune functions. But there are scanty reports regarding its effect on lipid profile in poultry. The study was conducted on 24 broiler chicks divided randomly into 3 groups, each group consisting of 8 birds. Group I birds were used as control kept on conventional diet. Group II and III birds were supplemented with garlic at the rate of 1.5 and 3.0%, respectively (on dry matter basis) of total feed for a period of 8 weeks. Lipid profile viz. total cholesterol, triglycerides, Low Density Lipoprotein (LDL), Very Low Density Lipoprotein (VLDL) and High Density Lipoprotein (HDL) were studied. The total cholesterol, triglycerides, LDL and VLDL were significantly decreased, while HDL was significantly increased by garlic supplementation in chicken up to 8 weeks of age in comparison to control group. There was a significant increase in total cholesterol with advancement of age and this increase was prevented by garlic supplementation in feed. The present findings suggest that the garlic supplementation in feed is effective in regulation of lipid metabolism, which is the predisposing factor for the coronary heart diseases. Further, our results suggest that the garlic is effective in regulation of cholesterol level with advancement of age. In conclusion, garlic is effective in regulation of lipid profile.

**Key words:** Broiler, cholesterol, garlic, lipid profile, triglycerides

**INTRODUCTION**

The virtues of garlic (*Allium sativum*), as a medicinal plant are known to most cultures of the world. Garlic is one of the oldest cultivated plant (Moyers, 1996). Garlic has been shown to have several effects in the body. These include inhibition of platelet aggregation (Apitz-Castro et al., 1983), reduction in arterial blood pressure (McMohan and Vargas, 1993) and prevention of fat infiltration of liver (Sand et al., 1995). Also, in vivo and in vitro studies showed that aged garlic extracts stimulate immune functions (Sumiyoshi, 1997). Previously, attention has also been focused on garlic's ability to lower cholesterol levels in humans (Adler and Holub, 1997) and animals (Aouadi et al., 2000). Also, Bordia et al. (1975) reported that garlic can prevent fat-induced hyperlipemia. Conversely, garlic failed to influence the lipid profiles in rats (Islam and Choi, 2008).

Broiler industry is a source of nutrition, provides rural employment and fulfills all nutritional standards. The success of poultry as industry in Indian scenario is evident from the fact that with 10 kg of similar feed, broiler type chicken gives 450 g of protein while swine gives 160 g, beef type cow gives 96 g and sheep and goat around 225 g of protein (Qureshi et al., 1983b). Therefore, chicken and eggs are inexpensive sources of protein among all animal products. In addition, depressed hepatic cholesterol levels in chicken were observed by Sklan et al. (1992). Suggesting that garlic supplementation can be of value in production of quality broiler meat.

In the light of lack of information on the influence of garlic on lipid profile parameters in chicken, this study was undertaken to investigate the influence of garlic on certain lipid profile parameters in chicken.

**MATERIALS AND METHODS**

Investigations were carried out on the 24, day old, unsexed and healthy commercial broiler chicks, procured from a commercial breeding farm. All the chicks belonged to the same batch and the same breeding stock. The chicks were reared under strict hygienic conditions. Before housing the chicks; rooms, brooder battery and cages were thoroughly cleaned with 2.5% phenol and subsequently fumigated with formaldehyde gas (35 mL of commercial formalin and 17.5 g potassium permanganate per hundred cubic feet area). Electric bulbs were used as source of heat and light. The experimental chicks were reared for eight weeks on the prepared feed procured from Department of Animal Nutrition, CCS Haryana Agricultural University, Hisar. Garlic purchased from the Department of Vegetable Crops, CCS Haryana Agricultural University Hisar, were crushed, shade dried and ground with mixer grinder to powdered form and was supplemented in the feed.
Experimental design: Twenty four, day old broiler chicks were kept for 1 week on chick mash. On the first day, chicks were divided into 3 groups i.e., Group-I, II and III, each consisting of 8 birds. Birds of Group-I, II and III were vaccinated against Newcastle Disease (ND) by F1 strain vaccine (intranasally) procured from Indovax Pvt. Ltd., India, on the 3rd day. Birds of Group-I kept as control and maintained on conventional poultry feed. Birds of Group-II were maintained on same feed along with garlic supplemented in feed at the rate of 1.5% (on dry matter basis) of total feed. Birds of Group-III were also maintained on same feed along with garlic supplemented at the rate of 3% (on dry matter basis) of total feed for a period of 8 weeks.

Collection of Blood Sample for Lipid Profile
Parameters: Blood samples from all the three groups were collected on 4, 6 and 8th week of age by cardiac puncture for lipid profile studies. Blood was collected in clean glass vials using dried heparin @ 15 IU/mL of blood as an anticoagulant. Plasma was separated and stored at -20EC till analysis.

Lipid profile studies
Total plasma cholesterol: Total cholesterol in plasma estimation was carried out by employing chemistry auto analyzing kit AUTOPAK® supplied by Bayer Diagnostics India Ltd, using enzymatic (Cholesterol Esterase, Cholesterol Oxidase and Peroxidase) method. The results are expressed as mg/dL.

Triglycerides in plasma: Triglycerides in plasma estimation was carried out by employing chemistry auto analyzing kit AUTOPAK® supplied by Bayer Diagnostics India Ltd using enzymatic (Lipoprotein lipase, Glycerol Kinase, Glycerol-3-Phosphate Oxidase, Peroxidase, 4-Aminoantipyrine and ATP) colorimetric method. The results are expressed as mg/dl of plasma.

HDL (High Density Lipoprotein)-Cholesterol in plasma: HDL-Cholesterol in plasma estimation was carried out by employing chemistry auto analyzing kit AUTOPAK® supplied by Bayer Diagnostics India Ltd using phosphotungstate method. The results are expressed as mg/dL of plasma.

VLDL (Very Low Density Lipoprotein)-Cholesterol in plasma: The plasma VLDL-Cholesterol was estimated by employing the Friedwald formula. The results are expressed in mg/dL of plasma.

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\text{VLDL-Chol} = \frac{\text{Plasma triglycerides}}{5}
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LDL (Low Density Lipoprotein)-Cholesterol in plasma: The plasma LDL-Cholesterol was estimated as the difference between total cholesterol and the sum of VLDL-Cholesterol and HDL-Cholesterol. The results are expressed in mg/dl of plasma.

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\text{LDL-Chol} = \text{Total Chol} - (\text{VLDL-Chol} + \text{HDL Chol})
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Statistical analysis: The data was statistically analyzed using ANOVA followed by t-test for comparison of them among different group using Sigma stat software.

RESULTS
Total cholesterol: The mean of total cholesterol in birds of different groups are depicted in Table 1. It revealed a significant (p ≤ 0.05) decrease in the level of total cholesterol in group II (144.2±3.16 mg/dL) and group III (136.4±3.53 mg/dL) in comparison to group I (155.8±2.87 mg/dL) at 4 weeks post treatment. On 6th week of post treatment there was significant (p < 0.05) decrease in total cholesterol concentration in group II (144.8±4.04 mg/dL) and group III (138.3±2.19 mg/dL) as compared to group I (184.1±4.47 mg/dL). Also, on 8th week of post treatment there was significant (p < 0.05) decrease in total cholesterol concentration in group II (147.8±2.99 mg/dL) and group III (139.5±3.60 mg/dL) as compared to group I (193.5±4.41 mg/dL). When the data (i.e., 4, 6 and 8th weeks) was pooled, it showed a significant (p ≤ 0.05) decrease in total cholesterol level in group II (145.6±1.11 mg/dL) and group III (138.0±0.90 mg/dL) in comparison to group I (177.8±11.32 mg/dL). There was a significant (p < 0.05) increase in total cholesterol at 6 and 8 week of age in comparison to 4 week of age only in group I. Garlic supplementation inhibited the increase in cholesterol level with advancement of age.

HDL-Cholesterol: The mean of HDL-Cholesterol in birds of different groups are depicted in Table 2. There was significant (p ≤ 0.05) increase in the level of HDL-Cholesterol in group II (41.08±1.03 mg/dL) and in group III (45.50±1.97 mg/dL) in comparison to group I (32.65±1.82 mg/dL) at 4 weeks post treatment. On 6th week of post treatment there was significant (p < 0.05) increase in HDL-Cholesterol concentration in group II (44.55±1.22 mg/dL) and group III (51.23±1.96) as compared to group I (34.40±1.83 mg/dL). Also, on 8th week of post treatment there was significant (p ≤ 0.05) increase in HDL-Cholesterol concentration in group II (50.01±2.91 mg/dL) and group III (50.60±1.46 mg/dL) in comparison to group I (32.18±1.47 mg/dL). When the data (i.e., 4, 6 and 8th weeks) was pooled, it observed that there was significant (p ≤ 0.05) increase in HDL-Cholesterol in group II (45.21±2.59 mg/dL) and in group III i.e. (49.11±1.81 mg/dL) in comparison to group I (33.07±0.67 mg/dL). However, with the advancement of age there was no significant (p ≤ 0.05) change in HDL-Cholesterol level in control group. But in garlic treated group II, it was a significant (P ≤ 0.05) increased at 8th
week of age in comparison to 4 and 6th week of age. While in group III, the increase was statistically insignificant.

**VLDL-Cholesterol:** The mean of VLDL-Cholesterol in birds of different groups are depicted in Table 3. There was a significant (p ≤ 0.05) decrease in the level of VLDL-Cholesterol in group II (17.43±0.62 mg/dL) and group III (14.77±0.88 mg/dL) in comparison to group I (19.39±0.60 mg/dL) at 4 weeks post treatment. On 6th week of post treatment there was significant (p ≤ 0.05) decrease in VLDL-Cholesterol concentration in group II (16.80±0.50 mg/dL) and group III (17.15±1.36 mg/dL) as compared to group I (21.92±0.66 mg/dL). Also, on 8th week of post treatment there was significant (p ≤ 0.05) decrease in VLDL-Cholesterol concentration in group II (17.10±1.53 mg/dL) and group III (15.33±1.97 mg/dL) as compared to group I (24.06±1.97 mg/dL). When the data (i.e., 4, 6 and 8th weeks) was pooled, it observed that there was significant (p ≤ 0.05) decrease in group II (17.11±0.18 mg/dL) and group III (15.75±0.71 mg/dL) in comparison to group I (21.79±1.34 mg/dL). However, with the advancement of age there was no significant (p ≤ 0.05) change in VLDL-Cholesterol level in groups I, II and III.

**LDL-Cholesterol:** The mean of LDL-Cholesterol in birds of different groups are depicted in Table 4. It showed a significant (p ≤ 0.05) decrease in the level of LDL-Cholesterol in group II (89.56±3.08 mg/dL) and group III (83.74±2.86 mg/dL) in comparison to group I (103.70±2.23 mg/dL) at 4 weeks post treatment. On 6th week of post treatment there was significant (p ≤ 0.05) decrease in LDL-Cholesterol concentration in group II (96.34±4.17 mg/dL) and group III (85.13±2.41 mg/dL) as compared to group I (127.23±4.12 mg/dL). Also, on 8th week of post treatment there was significant (p ≤ 0.05) decrease in LDL-Cholesterol concentration in group II (91.03±3.55 mg/dL) and group III (77.10±6.08 mg/dL) as compared to group I (137.21±3.02 mg/dL). When the data (i.e., 4, 6 and 8th weeks) was pooled, it showed a significant (p ≤ 0.05) decrease in group II (92.31±2.05 mg/dL) and group III (81.88±2.44 mg/dL) in comparison to group I (122.7±9.91 mg/dL). However, with the advancement of age there was no significant (p ≤ 0.05) change in LDL-Cholesterol level in groups II and III. But in group I, there was significant (p ≤ 0.05) increase in LDL-Cholesterol level at the age of 6 and 8th week of post treatment.

**Triglycerides:** The mean of triglycerides in birds of different groups are depicted in Table 5. There was a significant (p ≤ 0.05) decrease in the level of triglycerides in group II (87.11±2.93 mg/dL) and group III (73.82±2.70 mg/dL) in comparison to group I (96.90±2.87 mg/dL) at 4 weeks of treatment. On 6th week of post treatment there was significant (p ≤ 0.05) decrease in triglycerides concentration in group II (84.03±3.34 mg/dL) and group III (85.75±4.32 mg/dL) as compared to group I.
(109.90±3.14 mg/dL). On 8th week of post treatment there was significant (p < 0.05) decrease in triglycerides concentration in group II (85.51±5.63 mg/dL) and group III (76.62±3.49 mg/dL) as compared to group I (120.32±5.25 mg/dL). When the data (i.e., 4, 6 and 8th weeks) was pooled, it showed a significant (p < 0.05) decrease in group II (85.5±0.88 mg/dL) and group III (78.77±3.59 mg/dL) in comparison to group I (108.9±6.76 mg/dL). However, with the advancement of age there was no significant (p < 0.05) change in triglycerides level in groups II and III except group I.

**DISCUSSION**

**Total cholesterol:** In the present study, dietary supplementation of garlic powder at both concentrations (i.e., 1.5 and 3.0%) in broiler chickens was found to cause a significant (p < 0.05) decrease in the mean values of total cholesterol as compared to control birds. There was no significant (p < 0.05) difference in mean values of plasma total cholesterol in group III (3.0% garlic) as compared to group II (1.5% garlic) at any given time of interval. There was a significant (p < 0.05) increase in total cholesterol at 6 and 8 week of age in comparison to 4 week of age only in group I. This may probably be due to the possible mechanism of hypocholesterolaemic and hypolipidemic action of garlic products which depresses the hepatic activities of lipogenic and cholesterogenic enzymes such as malic enzyme, fatty acid synthase, glucose-6-phosphatase dehydrogenase (Chi et al., 1982; Qureshi et al., 1983a) and 3-hydroxy-3-methyl-glutaryl-CoA (HMG-CoA) reductase (Qureshi et al., 1983b, 1987). Afzal et al. (1985) reported that polyunsaturated fatty acids prevent atherosclerosis through the formation of cholesterol esters. They further reported the presence of higher polyunsaturated fatty acids like arachidonate and eicosapentanoate in garlic which could well be responsible for preventing atherosclerosis.

There are several reports in literature regarding the effect of garlic products administration on total cholesterol value in many species as well as poultry. Augusti (1977) reported that treatment with aqueous extract of garlic to hypercholesterolaemic human patients for 2 months (dose 0.5 mL/kg/day) significantly reduces the cholesterol levels. He ascribed this property of garlic to its sulphur containing compounds which can react with -SH group systems. Sodimu et al. (1984) revealed that administration of garlic oil (100 mg/kg b.Wt. /day) for one month together with HF-HC diet to another group of rats almost nullified the lipid-increasing and albumin decreasing effects of that diet. Powdered garlic significantly reduces cholesterol and triglyceride in blood (Grunwald et al., 1993).

Allicin is a specific inhibitor of the acetyl CoA synthetase enzyme necessary for fatty acid biosynthesis. Binding of allicin to the enzyme is non-covalent and reversible. Inhibition of fatty acid and lipid formation, by allicin may be a basis for use of allicin and garlic in folk medicine for cardiovascular diseases (Focke et al., 1990). Studies in Japanese quails indicated that dietary onion and garlic causes an insignificant decrease in plasma Esterified Cholesterol (EC) levels (Girish Kumar et al., 1998). Further, Konjufca et al. (1997) reported that the supplementation of 1.5% garlic in feed is enough to reduce plasma total cholesterol. Addition of 3.0 or 4.5% garlic powder does not further affect plasma cholesterol levels. Eidi et al. (2006) reported that oral administrations of garlic extract (0.10, 0.25 and 0.50 g/kg body wt.) for 14 days in diabetic rats significantly decreased total cholesterol, triglycerides, while increased serum insulin in diabetic rats but not in normal rats (p < 0.05). It is permissible to draw conclusions from the few avian species investigated to the entire class of Aves, then it becomes clear that serum or plasma cholesterol of birds is strongly affected by heredity, nutrition, age, sex and environment conditions (Sturkie, 1986). This significant increase in total plasma cholesterol level in this study may be the effect of age in poultry. This rise in cholesterol with age is prevented by garlic supplementation in feed. Conversely, garlic failed to influence the lipid profiles in rats (Islam and Choi, 2008). In view of the above findings and the present study it is suggested that garlic has cholesterol lowering effect in broiler chickens.

**HDL-Cholesterol:** In this study, dietary supplementation of garlic powder in groups i.e., I and III was found to cause a significant (p < 0.05) increase in the mean values of HDL-Cholesterol (HDL-C) as compared to group I. There was no significant (p < 0.05) difference in mean values of plasma HDL-C in group III as compared to group II at any given time of interval. However, with the advancement of age there was no significant (p < 0.05) change in HDL-C level but in garlic treated groups (i.e., II and III) except 8th week of age in group II, otherwise there was a non significant (p < 0.05) increase in HDL-C level in group II and III. This may probably be due to the possible mechanism of hypocholesterolaemic and hypolipidemic action of garlic products. Allicin, the
component of garlic, combining with the -SH group inactivate CoA, which is necessary for the biosynthesis of cholesterol. Also, allicin is a specific inhibitor of the acetyl CoA synthetase enzyme necessary for fatty acid biosynthesis. Binding of allicin to the enzyme is non-covalent and reversible in nature. Inhibition of fatty acid and lipid formation, by allicin may be a basis for hypocholesterolaemic and hypolipidemic action of garlic (Focke et al., 1990). Further, HDL-C levels are inversely proportional to the circulating cholesterol level. There are reports in literature regarding the effect of garlic products administration on HDL-C value in many species as well as poultry. Reports indicate inconsistent variations in the plasma HDL-C level due to dietary onion and garlic in Japanese quails (Augusti and Mathew, 1973; Girish Kumar et al., 1998). Similarly, inconsistent effect due to feeding these alliums on HDL-C was obtained in rats, broilers and layers (Chi et al., 1982; Qureshi et al., 1983a). Therefore, the present study is in accordance with the various reports available in the literature indicating that garlic supplementation in feed increases HDL-C in broiler chickens. There are no reports in literature on HDL-C in comparison with the advancement of age. This indicates that garlic products or its bioactive metabolites increase HDL-C with the advancement of age as the results of this study suggests.

VLDL-Cholesterol: The dietary supplementation of garlic powder in groups II and III was found to cause a significant ($p \leq 0.05$) decrease in the mean values of VLDL-Cholesterol (VLDL-C) as compared to group I. However, there was not any significant ($p \leq 0.05$) difference in mean values of plasma VLDL-C in group III as compared to group II at any given time of interval. Also, with the advancement of age there was no significant ($p \leq 0.05$) change in VLDL-C level in groups I, II and III. The possible mechanism of lowering in VLDL-C by garlic products is unclear. But probably it may due to increase in the esterified cholesterol by garlic supplementation may be due to either decreasing the production or increasing the excretion of esterified cholesterol through the bile. This may lead to decrease in VLDL-Cholesterol in garlic supplemented groups (Girish kumar et al., 1998).

There are reports in literature regarding the effect of garlic products administration on VLDL-C value in many species as well as poultry. Augusti and Mathew (1973) reported a better transport and utilization of lipids by the tissues under the influence of alliums (components of garlic). Diet plays an important role in modulating plasma cholesterol. Excessive energy intakes increase hepatic production of VLDL-C. The transport system of cholesterol from liver to tissues is VLDL-C and the utilization of cholesterol is by LDL-C, the precursor for LDL-C is again VLDL-C (Grundy, 1986). Studies indicated that dietary onion and garlic causes an insignificant decrease in plasma Esterified Cholesterol (EC) levels (Augusti and Mathew, 1973). Similarly, inconsistent effect due to feeding these alliums on VLDL-C was obtained in rats, broilers and layers (Chi et al., 1982; Qureshi et al., 1983a). There are no reports in literature on VLDL-C in comparison with the advancement of age. This indicates that garlic products or its bioactive metabolites decrease VLDL-C with the advancement of age as the results of this work suggests.

LDL-Cholesterol: Garlic supplementation in groups II and III was found to cause a significant ($p \leq 0.05$) decrease in the mean values of LDL-Cholesterol (LDL-C) as compared to group I. However, there is no significant ($p \leq 0.05$) difference in mean values of plasma LDL-C in group III as compared to group II at any given time of interval. Also, with the advancement of age there was no significant ($p \leq 0.05$) change in LDL-C level in groups II and III. But in group I, there was significant ($p<0.05$) increase in LDL-C level at the age of 6 and 8th week of post treatment. This may probably be due to the possible mechanism of antioxidant and antiperoxide lowering action of garlic products i.e., S-allyl Cysteine Sulfoxide (SAC) on LDL-C or decrease in hepatic production of VLDL-C which serves as the precursor of LDL-C in the blood circulation (Grundy, 1986). There are reports in literature regarding the effect of garlic products administration on LDL-C value in many species as well as poultry. The decrease in LDL-C level due to feeding these alliums on LDL-C was obtained in rats, broilers and layers (Chi et al., 1982; Qureshi et al., 1983a). Girish Kumar et al. (1998) reported decrease in the level of plasma LDL-C in both sexes due to dietary supplementation of onion and garlic supplementation in Japanese quails. There are no reports in literature on LDL-C in comparison with the advancement of age. Thus, as this study indicates that garlic products or its bioactive metabolites decrease LDL-C with the advancement of age.

Triglycerides: Dietary supplementation of garlic powder was found to cause a significant ($p \leq 0.05$) decrease in the mean values of triglycerides as compared to control group. However, no significant ($p \leq 0.05$) difference in mean values of plasma triglycerides in group III as compared to group II at any given time of interval. Also, with the advancement of age there was no significant ($p \leq 0.05$) change in triglycerides level in groups II and III except group I. The change in triglycerides level due to garlic supplementation might probably be due to the possible mechanism of allicin, the organosulfur component of garlic, combining with the -SH group (Cavallito et al., 1944), the functional part of CoA, which is necessary for the biosynthesis of triglyceride. Also,
allicin is a specific inhibitor of the acetyl CoA synthetase enzyme necessary for fatty acid biosynthesis. Binding of allicin to the enzyme is non-covalent and reversible in nature. Inhibition of fatty acid and lipid formation, by allicin may be a basis for hypocholesterolaemic and hypolipidemic action of garlic (Focke et al., 1990). This may be the cause of decreased triglycerides level in lard. J. Nutr., 112: 241-248.

Hypolipidemic action of garlic (Focke et al., 1990). This garlic on lipid metabolism in rats fed cholesterol or lard. J. Nutr., 112: 241-248.


