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# Influence of Dietary Mixtures of Garlic and Ginger on Lipid Composition in Serum, Yolk, Performance of Pullet Growers and Laying Hens

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Abstract: Medicinal spices such as garlic and ginger contain chemical substances that could be used to enhance the value of food of animal origin. These substances such as organosulphur compounds of garlic and gingerone, shoagaols of ginger are responsible health benefits of these spices. The study investigated the addition of mixtures of garlic and ginger to diets of pullet chicks, growers and laying hens to evaluate beneficial effects on growth and egg production as well as their possible effects on lipid content of serum and volk. Five diets were used for the study. Control diet was not supplemented with any of the mixtures. Four increasing levels of garlic (0.5%, 1.0%, 1.5% and 2.0%) were incorporated into diets for pullet chicks, growers and laying hens. Ginger supplements were also added to the diets at 0.5% and 0.75%. The results showed that the mixtures of garlic and ginger were beneficial by promoting final live weight of pullet growers, laying hens and also by lowering total cholesterol, triacylglycerol concentration in serum and yolk. Furthermore, the mixtures of garlic and ginger significantly (p<0.001) improved hen day production, egg weight and feed conversion. However, dietary inclusion of the mixtures significantly reduced shell weight and thickness. Significant (p<0.001) negative linear relationships were observed between dietary mixtures of garlic and ginger with parameters of serum and yolk lipids. In conclusion, the mixtures of garlic and ginger improved egg weight with lower cholesterol and triacylglycerol as well as better hen day production of laying hens. The results implied that the mixtures could benefit growth, laying performance and lowered yolk lipid content.

Key words: Hens, yolk, cholesterol, garlic and ginger mixtures

#### INTRODUCTION

The significant contribution of dietary fat in human foods to development of chronic and terminal diseases in human beings such as cardiovascular diseases and cancer has been elaborated in many studies. Foods that contain high saturated fatty acids and cholesterol are risk factors to the incidence of cardiovascular diseases (Grundy, 1986; John et al., 2009), diabetes (Villegas et al., 2004) and colon cancer in human (Mckeown-Eyssen and Bright-See, 1984; Willet et al., 1990; Boyd et al., 2003). The fat content and the composition of fatty acids in egg lipids have been implicated in human health (British Nutrition Foundation, 1992; Chow, 1992). Intake of saturated fatty acids had a direct relationship with the incidence of cardiovascular diseases (Norum, 1992). Furthermore, Taylor et al. (2007) concluded that women, both pre- and post-menopausal, who consumed meat had the highest risk of breast cancer. Processed meat showed the strongest Hazard ratio of 1.64 (95% CI: 1.14-2.37) for consumption than those who did not consumed meat.

The growing appreciation for low fat poultry products by consumers has encouraged researches to be focused in lowering cholesterol and triacylglycerol of poultry products using many agents such as copper sources. However, reduced egg production (Ankari et al., 1998), its negative effects in egg weight (Mabe et al., 2003) and feed conversion (Ankari et al., 1998), as well as its associated economic loss have cast doubt in using these agents for a profit oriented egg laying enterprise. Many medicinal plants such as garlic, ginger possess health promoting organic substances such as organosulphur compounds of garlic (Amagase et al., 2001), gingerone and shoagaol of ginger (Fuhrman et al., 2000) that may be used in lowering yolk cholesterol and triacylglycerol.

Birrenkott *et al.* (2000) reported that 3% garlic powder did not have significant effect on yolk and serum cholesterol concentrations when laying hens fed diets for 8 months. However, Khan *et al.* (2007) reported that dietary garlic powder at 2, 6 and 8% did not significantly affect feed intake, feed efficiency, egg weight, egg mass,

but significantly reduced serum and yolk cholesterol. Ademola *et al.* (2009) reported that mixtures of garlic and ginger in broiler diet enabled broiler chicks and chickens to achieve maximum final live weights as well as effectively reduced abdominal fat, serum cholesterol and triacylglycerol than sole dietary garlic and ginger. The positive influence of the mixtures of garlic and ginger on growth and lipid metabolism of broilers stimulated the current investigation. The study therefore examined growth, serum lipids of pullet birds, yolk lipid content, performance of laying hens fed different mixtures of garlic and ginger.

# **MATERIALS AND METHODS**

**Sources of test ingredients:** Garlic and dried ginger were obtained from a local market in Ogbomoso, Nigeria.

Processing of test ingredients: Garlic bulbs were carefully desegmented into cloves. These cloves were descaled while each clove was sliced into small chips with knife for effective drying. Garlic chips were sundried for 2 to 3 weeks at environmental temperature of 38.83±1.87°C. Sun dried garlic chips were stored in a plastic container prior to the study. Dried ginger were pounded and milled. Thereafter, milled ginger was packed in an airtight polythene sack.

**Experimental diets:** Five diets were prepared for pullet chick, grower and layer experiments. Pullet chick, grower and layer diets contained similar levels of mixtures of garlic and ginger. Control diet was not supplemented with the mixture of garlic and ginger. Four increasing levels of garlic (0.5%, 1.0%, 1.5% and 2.0%) were incorporated into diets for pullet chicks, growers and laying hens labeled as T1, T2, T3 and T4 respectively. Ginger supplements were added for diets in T1, T2 at

0.5%, while 0.75% ginger was included in diets T3 and T4. The arrangement led to inclusion of four different mixtures of garlic and ginger in the experimental diets. Pullet chicks diets and grower diets contained 2792.91± 21.2 kcal metabolizable energy (ME)/kg, 19.7±0.19% Crude Protein (CP) and 2525.53±21.6 kcal ME/kg, 16.1±0.08% CP respectively. The gross composition of layer diet is as shown in Table 1.

# Management of pullet birds and laying hens

Experiment 1: Three hundred day old ISA Brown pullet chicks were randomly distributed into five dietary treatments. Each treatment has four replicates of 60 chicks. Pullet chick diets were offered to the chicks from day old to 5th weeks of age, while grower diets were offered to the birds from 6th weeks to 22nd week of age until the birds dropped the first egg. Fresh feeds and water were served ad libitum daily. Routine management and all necessary vaccination procedures were followed. The birds were transferred from deep litter system to battery cage system at 14th week of age. This portion of the study was carried out in October, 2005 from day old to 22 weeks.

**Experiment II:** A total of selected 240 point of lay hens of ISA Brown strain were randomly distributed to five dietary treatments as earlier described in experiment I. Each treatment contained 48 hens of 4 replicates. The hens were housed individually in a battery cage for 32 weeks. The hens were offered feed and clean water *ad libitum* throughout the trial. Yolk colour was determined by Roche yolk fan. The study was carried out immediately after experiment I for 32 weeks in 2006.

Chemical analysis: On the 18th and 40th week of age of experimental hens, 3 blood samples per replicate were collected without anticoagulant for estimation of serum

| Table 1: Ingredien | composition of la   | yer diets containing | mivtures of      | gadic and ginger |
|--------------------|---------------------|----------------------|------------------|------------------|
| Table L. Higheuleh | i composition of la | yei ulets containing | I IIIIXIUI ES UI | ganic and ginger |

| Ingredients                  | Control | T1      | T2      | Т3      | T4      |
|------------------------------|---------|---------|---------|---------|---------|
| Maize                        | 44.28   | 44.28   | 44.28   | 44.28   | 44.28   |
| Soya bean meal               | 12.92   | 12.92   | 12.92   | 12.92   | 12.92   |
| Palm kernel meal             | 8.00    | 7.70    | 7.55    | 7.33    | 7.18    |
| Maize bran                   | 7.00    | 6.70    | 6.55    | 6.32    | 6.90    |
| Wheat bran                   | 8.00    | 7.60    | 7.40    | 7.10    | 6.90    |
| Fixed ingredients*           | 19.80   | 19.80   | 19.80   | 19.80   | 19.80   |
| Garlic                       | -       | 0.50    | 1.00    | 1.50    | 2.00    |
| Ginger                       | -       | 0.50    | 0.50    | 0.75    | 0.75    |
| Nutrient composition         |         |         |         |         |         |
| Calculated energy (kcal/kg)  | 2610.02 | 2588.52 | 2576.67 | 2561.61 | 2550.86 |
| Determined crude protein (%) | 17.45   | 17.30   | 17.62   | 17.53   | 17.71   |
| Determined calcium (%)       | 3.55    | 3.52    | 3.51    | 3.51    | 3.50    |
| Determined phosphorus (%)    | 0.45    | 0.45    | 0.45    | 0.45    | 0.44    |

<sup>\*</sup>Fixed ingredient contained 7.00% groundnut cake, 2.00% fish meal, 3.00% bone meal, 7.00% oyster shell, 0.25% salt, 0.25% vitamin\*, 0.15% methionine and 0.15% lysine.

<sup>\*</sup>Layer vitamin premix supplied the following vitamins and trace elements per kg diet: Vit A 6250 IU; VitD3 1250IU; VitE, 14.38 mg; Vitk3 1.25 mg, VitB, 1.88 mg Vit B2 3.75 mg; Niacin 31.25 mg; calcium pantothenate 6.25 mg; Vigt B6 3.8 mg, Vit B12 0.02 mg; Choline Chloride 250 mg; Folic acid 0.63 mg; Biotin 0.03 mg; Mn 75 mg; Fe 62.5 mg; Zn 50 mg; Cu 5.31 mg; I 0.94 mg; Co 0.019 mg; Se 0.08 mg and Antioxidant 75 mg

lipids of the hens. Serum total cholesterol, triacylglycerol and High Density Lipoprotein (HDL) cholesterol were determined using sigma enzymatic kits according to the methods of Roeschlau *et al.* (1974), Wahlefeld (1976) and Warnick *et al.* (1982) respectively. Serum Low Density Lipoprotein (LDL) cholesterol was calculated (Friedewald *et al.*, 1972).

Determination of yolk cholesterol and triacylglycerol: Yolk cholesterol and triacylglycerol were determined by using Sigma enzymatic kits.

Statistical analysis: All data collected were analyzed by one-way analysis of variance of SPSS version 10. Significant means were separated using Duncan option of the same statistical package. Linear regressions of data collected from serum and yolk lipid contents were tested against dietary concentration of garlic and ginger mixtures with the same statistical package.

## **RESULTS**

The results of growth performance of pullet growers and final live weight of laying hens are shown in Table 2. Feeding mixtures of garlic and ginger significantly (p<0.01) influenced Final Live weight (FLW) and weight gain of pullet growers in experiment I. Growers fed mixtures of garlic and ginger in T2 had heaviest FLW while pullet growers in control had lowest FLW. Furthermore, laying hens fed mixture of garlic and ginger in T1 in experiment II had the heaviest FLW.

Serum lipid of pullet growers and laying hens are shown in Table 3. The mixtures of garlic and ginger significantly decreased serum total cholesterol, triacylglycerol and low density lipoprotein cholesterol of growers and laying hens. Pullet growers in T3 had the lowest serum total-and LDL-cholesterol concentrations. The mixtures of garlic and ginger also significantly lowered yolk cholesterol and triacylglycerol (Table 4). Laying hens in T2 had lowest serum total cholesterol and triacylglycerol. Furthermore, hens in T4 had lowest yolk cholesterol and triaylglycerol.

All the mixtures significantly (p<0.001) enhanced laying performance with hens in T2 having the best hen day production, superior feed conversion and heaviest egg weight (Table 4). Laying hens had similar weights of albumen and yolk. However, dietary inclusion of the mixtures significantly (p<0.001) reduced shell weight and shell thickness. Dietary garlic and ginger mixtures did not significantly affect yolk colour.

The linear regression equations of lipid content in the blood of pullet growers, laying hens and yolk are shown in Table 5. All the parameters showed significant (p<0.001) negative linear relationships with the mixtures of garlic and ginger except HDL cholesterol that has a positive linear relationship. However, the coefficients of determinations of the data collected were low. The mixtures established the highest correlation with serum LDL cholesterol of pullet growers and lowest relationship with serum triacylglycerol of laying hens.

Table 2: Growth performance of 22-week pullet growers (Expt I) and final live weight of 54-week old laying hens (Expt II) fed mixtures of garlic and ginger (g/bird)

| Parameters                                   | Control   | T1                    | T2        | T3        | T4                    | SEM                |
|--|-----------|-----------------------|-----------|-----------|-----------------------|--------------------|
| Grower final live weight                     | 1270.33°  | 1288.86 <sup>bc</sup> | 1360.21ª  | 1331.73ab | 1288.86 <sup>bc</sup> | 15.73**            |
| Initial li∨e weight                          | 34.50     | 34.50                 | 33.25     | 33.75     | 34.50                 | 0.64 <sup>NS</sup> |
| Grower weight gain (g/bird/day)              | 8.02⁵     | 8.15 <sup>bc</sup>    | 8.62ª     | 8.43ab    | 8.15 <sup>bc</sup>    | 0.10**             |
| Grower feed intake (g/bird/day)              | 53.12     | 48.61                 | 57.85     | 55.10     | 59.50                 | 4.00 <sup>NS</sup> |
| Feed conversion                              | 6.62      | 5.97                  | 6.49      | 6.52      | 7.31                  | 0.47 <sup>NS</sup> |
| Final live weight of 54 week old laying hens | 1610.28bc | 1695.00°              | 1638.93ab | 1556.32°  | 1664.72ab             | 24.33**            |

Means in the same row with different superscripts are significant different (\*\*p<0.01).

NS - Not significant different

Table 3: Serum lipid of pullet growers (Expt I) and laying hens (ExptII) fed garlic and ginger mixtures (mg/dl)

| Parameters        | Control            | T1                 | T2                  | T3                   | T4                  | SEM     |
|-------------------|--------------------|--------------------|---------------------|----------------------|---------------------|---------|
| Total cholesterol |                    |                    |                     |                      |                     |         |
| Pullet growers    | 108.17ª            | 90.22°             | 89.52 <sup>cd</sup> | 84.16 <sup>d</sup>   | 94.12ª              | 1.24*** |
| Laying hens       | 167.94ª            | 145.23b            | 93.94 <sup>d</sup>  | 104.68 <sup>cd</sup> | 113.49°             | 4.58*** |
| Triacylglycerol   |                    |                    |                     |                      |                     |         |
| Pullet growers    | 148.60°            | 89.83bc            | 95.83bc             | 91.52 <sup>bc</sup>  | 78.93⁰              | 4.35*** |
| Laying hens       | 179.44°            | 177.65°            | 147.06⁵             | 164.13⁵              | 178.23°             | 3.40*** |
| LDL cholesterol   |                    |                    |                     |                      |                     |         |
| Pullet growers    | 27.21ª             | 15.62 <sup>b</sup> | 14.83b              | 11.53 <sup>□</sup>   | 13.42 <sup>cd</sup> | 0.81*** |
| Laying hens       | 83.13°             | 41.02°             | 55.93⁵              | 50.00 <sup>b</sup>   | 37.84⁵              | 2.43*** |
| HDL cholesterol   |                    |                    |                     |                      |                     |         |
| Pullet growers    | 50.10 <sup>d</sup> | 58.07 <sup>b</sup> | 56.04 <sup>bc</sup> | 52.86 <sup>cd</sup>  | 62.73°              | 1.35*** |
| Laying hens       | 44.17⁵             | 44.25 <sup>b</sup> | 44.14 <sup>b</sup>  | 38.18 <sup>b</sup>   | 53.51°              | 3.40*** |

Means in the same row with different superscript are significant different (\*\*\*p<0.001)

Table 4: Laying performance, yolk lipid, egg quality of hens fed garlic and ginger mixtures

|                              | 1 / 00 1 /         |                     |                     |                     |                     |                    |
|------------------------------|--------------------|---------------------|---------------------|---------------------|---------------------|--------------------|
| Parameters                   | Control            | T1                  | T2                  | T3                  | T4                  | SEM                |
| Hen day prod (%)             | 60.58 <sup>d</sup> | 68.97 <sup>ab</sup> | 71.47ª              | 66.79 <sup>bc</sup> | 64.67⁰              | 1.09***            |
| Feed intake (g/bird/day)     | 116.15ª            | 115.91ª             | 118.28ª             | 112.03b             | 111.61 <sup>b</sup> | 1.56**             |
| Feed conversion (feed/egg)   | 3.39°              | 3.06°               | 3.05⁵               | 3.11 <sup>b</sup>   | 3.30°               | 0.05***            |
| Egg weight (g/egg)           | 55.36⁵             | 54.91⁵              | 58.14ª              | 56.75₺              | 56.50⁰              | 0.32***            |
| Yolk cholesterol (mg/ml)     | 11.26ª             | 10.54 <sup>ab</sup> | 8.67⁰               | 9.96⁰               | 8.24⁵               | 0.41***            |
| Yolk triacylglycerol (mg/ml) | 25.71 <sup>a</sup> | 20.14 <sup>b</sup>  | 19.93⁵              | 20.69⁵              | 19.51 <sup>b</sup>  | 0.76***            |
| Yolk weight (%)              | 24.28⁵             | 24.37⁵              | 25.08ab             | 24.37b              | 24.28b              | 0.02**             |
| Albumen weight (%)           | 62.84ab            | 63.24°              | 62.24b              | 63.19ª              | 63.25°              | 0.23**             |
| Shell + membrane (%)         | 12.88 <sup>a</sup> | 12.39°              | 12.68 <sup>ab</sup> | 12.44 <sup>bc</sup> | 12.47 <sup>bc</sup> | 0.09***            |
| Shell thickness (x10-02 mm)  | 33.12⁵             | 32.43⁵              | 33.52°              | 32.46°              | 33.06 <sup>b</sup>  | 0.13***            |
| Yolk colour                  | 1.00               | 1.01                | 1.03                | 1.01                | 1.01                | 0.01 <sup>NS</sup> |

Means in the same row with different superscripts are significantly different (\*\*p<0.01, \*\*\*p<0.001). NS - Not significantly different

Table 5: Linear regression equations of lipid content in the serum of pullet growers and yolk

| Parameters           | Linear equation | R    | R2   | SL  |
|----------------------|-----------------|------|------|-----|
| Total cholesterol    |                 |      |      |     |
| Pullet growers       | Y=103.49-3.42x  | 0.53 | 0.28 | *** |
| Laying hens          | Y=169.89-14.95x | 0.67 | 0.45 | *** |
| Triacylglycerol      |                 |      |      |     |
| Pullet growers       | Y=142.24-13.77x | 0.69 | 0.47 | *** |
| Laying hens          | Y=173.55-1.42x  | 0.12 | 0.01 | NS  |
| LDL cholesterol      |                 |      |      |     |
| Pullet growers       | Y=26.02-3.17x   | 0.73 | 0.53 | *** |
| Laying hens          | Y=78.06-8.16x   | 0.64 | 0.41 | *** |
| HDL cholesterol      |                 |      |      |     |
| Pullet growers       | Y=49.95+2.00x   | 0.45 | 0.21 | *** |
| Laying hens          | Y=40.96+1.36x   | 0.25 | 0.06 | NS  |
| Yolk cholesterol     | Y=11.72-0.66x   | 0.53 | 0.28 | *** |
| Yolk triacylglycerol | Y=24.75-1.19x   | 0.49 | 0.24 | *** |

Means in the same row with different superscripts are significantly different (\*\*\*p<0.001). NS - Not significantly different. SL = Significant Level

## DISCUSSION

The mixtures of garlic and ginger displayed performance enhancing effects as demonstrated in the growth rate of pullet growers, final live weight, hen day production and egg weight of laying hens. Ahmed and Sharma (1997) reported that the growth rate of rats fed mixture of garlic (2%) and ginger (0.5%) was significantly higher than those fed sole garlic and ginger. It had earlier been reported that the mixtures of garlic and ginger significantly promoted growth of broiler chicks by enhancing digestion of food nutrients and probably improved food absorption through the wall of gastrointestinal tract (Ademola et al., 2007). The mixtures of garlic and ginger significantly lowered total cholesterol, triacylglycerol in the blood of experimental birds and yolk. The hypolipidemic effects of garlic on yolk cholesterol and triacylglycerol have been reported (Reddy et al., 1991; Enyinna, 1995; Khan et al., 2007). Oven dried garlic did not significantly reduce yolk cholesterol but effectively lowered yolk triacylglycerol (Enyinna, 1995). Processing methods for garlic and the combined influence of garlic and ginger in this study may account for the variation in yolk cholesterol.

Intakes of differently processed garlic such as raw, frozen, dried by birds (Augusti and Mathew, 1993; Qureshi et al., 1983)), rats (Shashikanth et al., 1986; Slowing et al., 2001) broiler chickens (Ademola et al., 2004) did not significantly affect growth performance. Enyinna (1995) reported that oven dried garlic did not affect egg weight, egg production but significantly reduced shell thickness. Furthermore, Khan et al. (2007) reported that dietary garlic powder at 2, 6 and 8% did not significantly affect feed intake, feed efficiency, egg weight, egg mass, but significantly reduced serum and yolk cholesterol. The synergistic effects of the chemical compounds in garlic and ginger may account for the improved growth, hen day production and egg weight in this study.

Eggs with low concentration of volk cholesterol and triacylglycerol in this study will encourage a wholesome consumption of eggs by consumers, as high levels of these lipids have been implicated in metabolic disorders. Dietary cholesterol and fat especially saturated fat cause plasma concentration of cholesterol to rise by down-regulating LDL receptor synthesis (Spady and Dietschy, 1988; Dietschy et al., 1993). Consequently, this may increase the risk of developing coronary heart disease and atherosclerosis in humans. The significant negative linear relationship between dietary concentration of the mixtures of garlic and ginger with the parameters of serum and yolk lipids implied that low dietary concentration of the mixtures of garlic and ginger could be used to lower lipids in the blood and yolk. The low coefficient of determination for yolk lipids in this study revealed that yolk cholesterol and triacylglycerol were difficult to be lowered because of the physiological mechanism in egg as they served as nutrients available for young embryo development into chicks. The incorporation of garlic and ginger mixtures in layer diets did not significantly affect yolk colour. This indicated that the pigmentation process of volk involving oxycarotenoids was not tampered with despite the dietary inclusion of the mixtures of garlic and ginger. The beneficial action of the mixtures of garlic and ginger have been reported to lower abdominal fat pad, serum

triacylglycerol, total-and Low Density Lipoprotein (LDL)-cholesterol of broiler chickens (Ademola *et al.*, 2007) thereby enhancing dietetic value of these dressed chickens.

Conclusively, dietary garlic and ginger mixtures significantly improved laying performance in terms of hen day production, egg weight, feed conversion, weight gain, final live weights of pullet growers and laying hens. Consequently, these mixtures produced improved laying performance with the expected increased income if incorporated into layer diet. Additional benefits of the mixtures showed that they could be used to lower concentration of cholesterol and triacylglycerol in blood and egg yolk. However, the mixtures of garlic and ginger adversely affected egg shell weight and thickness.

#### **REFERENCES**

- Ademola, S.G., G.O. Farinu and G.M. Babatunde, 2009. Serum lipid, growth and haematological parameters of broilers fed garlic, ginger and their mixtures. World J. Agric. Sci., 5: 99-104.
- Ademola, S.G., G.O. Farinu, O.O. Adelowo, T.E. Lawal and G.M. Babatunde, 2007. Antimicrobial activity of garlic and ginger mixtures, serum lipid profile and growth performances of broilers fed the mixtures. Bowen J. Agric., 4: 103-113.
- Ademola, S.G., G.O. Farinu, A.O. Ajayi-Obe and G.M. Babatunde, 2004. Growth, haematological and biochemical studies on garlic-and ginger-fed broiler chickens. MOOR J. Agric. Res., 5: 122-128.
- Ahmed, R.S. and S.B. Sharma, 1997. Biochemical studies on combined effects of garlic (*Allium sativum* Linn) and ginger (*Zingiber offcinale* Roscoe) in albino rats. Indian J. Exp. Biol., 35: 841-843.
- Amagase, H., B. Petesch, H. Matsuura, S. Kasuga and Y. Itakura, 2001. Intake of garlic and bioactive components. J. Nutr., 131: 955S-962S.
- Ankari, A.Al., H. Najib and A.Al. Hozab, 1998. Yolk and serum cholesterol and production traits, as affected by incorporation a supraoptimal amount of copper in the diet of the leghorn hen. Br. Poult. Sci., 39: 393-397.
- Augusti, K.T. and P.T. Mathew, 1993. Effect of long-term feeding of the aqueous extracts of onion (*Allium cepa* Linn.) and garlic (*Allium sativum* Linn.) on normal rats. Indian J. Exp. Biol., 11: 239-241.
- Birrenkott, G., G.E. Brockenfelt, N. Owen and E. Halpin, 2000. Yolk and blood cholesterol levels and organoleptic assessment of eggs from hens fed garlic supplemented diet. Poult. Sci., 79: 75.
- Boyd, N.F., J. Stone, K.N. Vogt, B.S. Connelly, L.J. Martin and S. Minkin, 2003. Dietary fat and breast cancer risk revisited: A meta-analysis of the published literature. Br. J. Cancer, 89: 1672-1685.

- British Nutrition Foundation, 1992. Unsaturated fatty acids: Nutritional and physiological significance the report of the British nutrition foundation's task force. Chapman and Hall, London, UK. http://www.abebooks.com/9780412457500/Unsaturated-Fatty-Acids-Nutritional-Physiological-0412457504/plp.
- Chow, K.C., 1992. Fatty Acids in Foods and Their Health Implications. Marcel Dekker, New York, ISBN-13: 9780824786229.
- Dietschy, J.M., L.A. Woollett and D.K. Spady, 1993. The interaction of dietary cholesterol and specific fatty acids in the regulation of LDL receptor activity and plasma LDL-cholesterol concentrations. Ann. N.Y. Acad. Sci., 676: 11-26.
- Enyinna, J.U., 1995. Serum and egg yolk cholesterol of laying chickens fed graded of cholestyramine, garlic and vegetable oils. Ph.D. Thesis, Dept. of Animal Sci., Univ. of Ibadan, Ibadan, Nigeria.
- 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.
- Fuhrman, B., M. Rosenblat, T. Hayek, R. Coleman and M. Aviram, 2000. Ginger extract consumption reduces plasma cholesterol, inhibits LDL oxidation and attenuates development of atherosclerosis in atherosclerosis in atherosclerosis in atherosclerosis in atherosclerotic apolipoprotein Edeficient mice. J. Nutr., 130: 1124-1131.
- Grundy, S.M., 1986. Cholesterol and coronary heart disease. A new era. J. Am. Med. Assoc., 256: 2849-2858.
- John, C., L. Thomas and S. Patrick, 2009. The European Society of Cardiology Textbook of Cardiovascular Medicine. Blackwell Publishing, pp. 257.
- Khan, S.H., R. Sardar and M.A. Anjum, 2007. Effects of dietary garlic on performance and serum and egg yolk cholesterol concentrations in laying hens. Asian J. Poult. Sci., 1: 22-27. ISSN: 1819-3609.
- Mabe, I., C. Rapp, M.M. Bain and Y. Nyss, 2003. Supplementation of a corn-soybean meal with manganese, copper and zinc from organic or organic sources improves eggshell quality in aged laying hens. Poult. Sci., 82: 1903-1913.
- Mckeown-Eyssen, G.E. and E. Bright-See, 1984. Dietary factors in colon cancer: International relationships. Nutr. Cancer, 6: 160-170.
- Norum, K.R., 1992. Dietary fat and blood lipids. Nutr. Rev., 50: 30-37.
- 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.

- Reddy, R.V., S.F. Lightsey and D.V. Maurice, 1991. Research note: Effect of feeding garlic oil on performance and egg yolk cholesterol concentration. Poult. Sci., 70: 2006-2009.
- Roeschlau, P., E. Bernt and W. Gruber, 1974. Enzymatic determination of total cholesterol in serum. Z. Klin. Chem. Klin. Biochem., 12: 226-226.
- Shashikanth, K.N., S.C. Basappa and V.S. Murthy, 1986. Effect of feeding raw and boiled garlic (*Allium sativum* L.) on the growth, caecal microflora and serum proteins of albino rats. Nutr. Reports Int., 33: 313-319.
- Slowing, K., P. Ganado, M. Sanz, E. Ruiz and T. Tejerina, 2001. Study of garlic extracts and fractions on cholesterol plasma levels and vascular reactivity in cholesterol-fed rats. J. Nutr., 131: 994S-999S.
- Spady, D.K. and J.M. Dietschy, 1988. Interaction of dietary cholesterol and triglycerides in the regulation of hepatic low density lipoprotein transport in the hamster. J. Clin. Investig., 81: 300-309.

- Taylor, E.F., V.J. Burley, D.C. Greenwood and J.E. Cade, 2007. Meat consumption and risk of breast cancer in the UK Women's Cohort Study. Br. J. Cancer, 96: 1139-1146.
- Villegas, R., A. Salim, A. Flynn and I.J. Perry, 2004. Prudent diet and the risk of insulin resistance. Nutr. Metab. Cardiovasc. Dis., 14: 334-343.
- Wahlefeld, A.W., 1976. Triacylglycerol Determination After Enzymatic Hydrolysis. In: Methods of Enzymatic Analysis. Bergmeyer, U.H. (Ed.). 8th Edn., N.Y. Academic Press Inc. New York, pp. 1831-1835.
- Warnick, G.R., J. Benderson and J.J. Albers, 1982. Dextran sulfate-Mg<sup>2+</sup> precipitation procedure for quantitation of high-density-lipoprotein cholesterol. Clin. Chem., 28: 1379-1388.
- Willet, W.C., M.J. Stampfer, G.A. Colditz, B.A. Rosner and F.E. Speizer, 1990. Relation of meat, fat and fiber intake to the risk of colon cancer in a prospective study among women. New. Engl. J. Med., 323: 1664-1672.