# Modulation of serum and liver triglyceride and abdominal fat pad weight by dietary garlic in male broilers

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**Abstract:** In poultry, the liver is the main organ for the synthesis of triglycerides (TG) and their secretion into the plasma. Secretion of TGs from the liver and their storage in adipose tissue of the abdominal cavity reduces the meat yield. There are controversial reports about the effects of garlic on the levels of TG in the serum and liver of poultry. In this study, different diets that contained various levels of garlic powder were assessed with regards to their effects on the levels of TG in serum and the liver, and on the abdominal fat pad. For this purpose, 144 3 male Ross broilers who were three weeks old were assigned into four groups of 36 birds each, and fed on diets that were supplemented with 1, 2 or 4% of garlic powder, or with a diet that was not supplemented with garlic (the control group). At the end of the fifth and seventh weeks of the study, 15 chicks of each group were bled and then sacrificed. The livers and abdominal fat pad were removed. Liver and serum TG were extracted and measured by spectrophotometry. The TG levels of serum and the abdominal fat pad were not affected by diet by the end of the fifth week, but diets that contained 2% and 4% garlic powder had a significantly decreased hepatic TG level in comparison with the control group (p<0.05). At the end of the seventh week, a significant decrease in serum and hepatic TG levels was found in the birds that were fed a diet that contained 2% and 4% garlic powder, respectively (p<0.05). Also, the diet with 4% garlic powder significantly decreased the weight of the abdominal fat pad in comparison with the control group (p<0.05). It is concluded that garlic powder has the potential to decrease the levels of TG in the serum and liver in broilers in a dose-dependent manner.

Keywords: triglyceride, abdominal fat, broiler, garlic.

### Introduction

Lipid storage, particularly in the abdominal cavity as fat pad, is a major concern in poultry breeding because it affects the meat yield (Hermier, 1997). In poultry, lipogenesis is very limited in adipocytes (Saadoun and Leclercq, 1987). On the other hand, commercial avian breeds are usually fed

**\*Corresponding author:** Shahriary\_a@scu.ac.ir Tel: +98(611)3330073 Fax: +98(611)3360807 diets that are poor in lipids. Therefore, this fat pad is mainly due to primary hepatic lipogenesis (Leveille *et al.*, 1975; Pullen *et al.*, 1990; Griffin *et al.*, 1992). Triglyceride (TG) storage in adipose tissue depends on plasma lipid that originates mainly from hepatic TG synthesis and secretion. In an attempt to manipulate this fat pad, the factors that influence the



synthesis and secretion of TG have been studied. Some reports describe effect of garlic in lowering the levels of TG in the liver and plasma in poultry. Qureshi *et al.* (1983a) showed that methanolic and aqueous extracts of fresh garlic suppressed liver and plasma TG in broilers through the inhibition of fatty acid synthesis (FAS). The same researchers (Qureshi *et al.*, 1983b) showed that polar fractions of garlic decreased FAS activity in White Leghorn pullets. Depression of hepatic, muscle and serum cholesterol and TG levels in chickens fed with diets that contained 1.5%, 3%, and 4.5% of garlic powder was also observed by Konjufca *et al.* (1997).

The present study was conducted to determine the simultaneous effect of garlic on the levels of hepatic and serum TG, and its correlation with fat pad size, which has not been addressed previously in the literature.

### **Materials and Methods**

All chemical materials, including hexane (Merck, 822280), isopropanol (Merck, k995), chloroform (Merck, 822265), methanol (Merck, 6008), acetyl acetone (Merck, 800023), cholestrine (Merck, 3670), meta-periodate (Merck, 6597), glacial acetic acid (Merck, 90056), sulfuric acid (Merck, 713), ammonium acetate (Merck, 115), sodium sulfate (Merck, 6645), natrium hydroxide (Merck, 6462) and kalium hydroxide (Merck, 5012) were purchased from Merck. Co (Tehran, Iran).

#### Experimental birds and diets

In this study, 144 male 3-wk-old Ross  $\times$  Ross broiler chicks were assigned into four groups of 36 birds each and fed with mash diets (based on corn and soybean) supplemented with 1%, 2% or 4% garlic powder. The control group was fed with the mash diet without garlic supplementation. The diets were formulated in accordance with the NRC recommendations (1994) to meet the nutritional requirements of broilers. The composition of the basal diet is shown in Table 1. The birds had free access to feed and water throughout the experiment.

Table 1: Composition	of the basal diet.
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Ingredients	Content (%)	Nutrients	Calculated Content
Yellow corn	63.15	Protein (%)	19.10
Soybean meal	28.50	ME (kcal/g)	3.09
Fish meal	2.50	Ca (%)	0.90
Soybean oil	2.50	Available P (%)	0.42
Oyster shell	1.70	Lysine (%)	1.05
Dicalcium phosphate	0.80	Arginine (%)	1.20
Vitamin premix <sup>1</sup>	0.25	Methionine (%)	0.42
Mineral premix <sup>2</sup>	0.25	Meth.+Cystine (%)	0.72
Iodized sodium chloride	0.15	Na (%)	0.15
DL-methionine	0.15	Cl (%)	0.17
Lysine	0.03		
Coccidiostat	0.02		

<sup>1</sup>Provided per kilogram of diet: vitamin A (retinyl palmitate), 9,000 IU; cholecalciferol, 2,000 IU; vitamin E (dl-α-tocopheryl acetate), 18 IU; vitamin  $k_3$ , 2 mg; thiamine, 1.7 mg; riboflavin, 6.6 mg; niacin, 10 mg; pyridoxine, 2.9 mg; folic acid, 1 mg; choline Cl, 250 mg; biotin, 100 µg; vitamin B<sub>12</sub>, 15 µg; ethoxyquin, 120 mg. <sup>2</sup> Provided in milligrams per kilogram of diet: Mn, 99; Zn, 84; Fe, 50; Cu, 10; I, 1.9; Sel, 0.2.

#### Preparation of garlic powder

High quality garlic bulbs were purchased from local markets. To prepare the garlic powder, the peeled cloves were cut into smaller pieces and dried in oven at temperatures of between 50-60°C.

#### Lipid analysis

At the end of the fifth and seventh weeks of age, 15 chicks of each group were bled and then sacrificed. The livers and abdominal fat pad were kept in -20°C for lipid analysis after weighing. Liver and serum TG were extracted and measured by spectrophotometry. The level of serum TG was measured by the method described by Mendez *et al.* (1975). Lipids were extracted from the livers by the modified method of Hara and Radin (1978). Briefly, 9 ml of extraction solution (hexane: isopropanol 3:2 v/v) was added to 0.5 g of liver and homogenized by



glass beads for 8 h at room temperature. After homogenization, the organic phase was separated by centrifugation at  $2000 \times g$  for 10 min, dehydrated by saturated sodium sulphate and finally used for the TG assay according to the method of Neri and Frings (1973).

### Statistical analysis

The mean values of serum and hepatic TG, and the weight of the abdominal fat pad, were compared between the treatment groups using a one-way analysis of variance (ANOVA) and the post hoc Tukey test. All statistical analyses were performed using Sigma Stat 2 Software (Copyright 1992-1995 Jandel Corporation). Significance was accepted at the level of p<0.05.

# Results

All of the diets that contained 1%, 2%, and 4% garlic powder had no effect on serum TG by the end of the fifth week (p>0.05); diets that contained 2% and 4% garlic powder significantly decreased (p<0.05) the level of hepatic TG in comparison with the control group (Table 2).

Table 3 shows that at the end of the seventh week, a significant decrease in serum and hepatic TG levels was found in the birds that were fed diets containing 2% and 4% garlic, respectively (p<0.05). A significant decrease (p<0.05) in the weight of the abdominal fat pad was found in the birds fed with a diet that contained 4% garlic powder (Table 3).

# Discussion

The present study showed that the content of hepatic TG decreased by the fifth week of age, although serum TG was not affected by the diet. On the other hand, both the level of serum and hepatic TG decreased by the seventh week. Such results indicate that the lipid-lowering effect of garlic occurs earlier in the serum than the liver. This may be due to time insufficiency for the effect of garlic on serum TG. The lipid-lowering effect of garlic in the present study is in agreement with the findings of Qureshi *et al.* (1983a) in male broilers, Qureshi *et al.* (1983b) in White Leghorn pullets, Bordia (1981) in

**Table 2:** The effects of diets that contain various levels of garlic powder on the triglyceride content of serum and the liver, and on the abdominal fat pad/body weight ratio (mean $\pm$ SD) of broilers at the 5th week of age (n=15);

Garlic powder concentration (%)	0	1	2	4
Serum TG (mg/dl)	77.26± 11.93ª	73.53±15.64ª	68.10±12.28ª	71.44±12.53ª
Liver TG (mg/g liver)	66.44±13.45ª	58.36±12.08 <sup>ab</sup>	55.42±9.29 <sup>b</sup>	53.12±10.29 <sup>b</sup>
AFP/BW	1.10±0.25ª	1.14±0.225ª	0.967±0.220ª	0.976±0.176ª

Non-similar superscripts in each row are significantly different (p<0.05)

**Table 3:** The effect of diets that contain various levels of garlic powder on the level of triglyceride in serum and liver, as well as the abdominal fat pad/body weight ratio (mean $\pm$ SD) of broilers at the 7<sup>th</sup> week of age (n=15).

Garlic powder concentration (%)		1	2	4
Serum TG (mg/dl)	97.27± 8.13ª	97.93±11.68ª	84.86±9.39 <sup>b</sup>	80.27±10.54 <sup>b</sup>
Liver TG (mg/g liver)	62.18±12.72ª	63.62±9.45ª	52.32±9.22 <sup>b</sup>	49.08±6.93 <sup>b</sup>
AFP/BW	1.74±0.44ª	1.50±0.338 <sup>ab</sup>	1.470±0.285 <sup>ab</sup>	1.360±0.230 <sup>b</sup>

Non-similar superscripts in each row are significantly different (p < 0.05)

humans, and Yeh and Yeh (1994) in hepatocyte cultures of rats. On the other hand, in contrast with our findings, Carijjo *et al.* (2005) and Kumar *et al.* (2003) reported that dietary garlic paste did not alter the levels of serum TG in poultry and Japanese quail, respectively. However, Qureshi *et al.* (1983b) concluded that hypotriglyceridemic effect of dietary garlic in pullets appears by the fifth week of age, whereas in this present study, fresh dietary garlic had no such effect. This difference may be due to the kind of garlic product that was consumed in the study.

There are various reports with regards to the nature of the lipid-lowering effect of garlic products. Yeh and Yeh (1994) showed that garlic reduced plasma TG by inhibiting hepatic TG synthesis in primary cultures of rats. Qureshi *et al.* (1983a) reported that ethanolic and aqueous extracts of garlic inhibit FAS activity in male broilers. On the other



hand, Lee *et al.* (2000) have revealed that fresh garlic extracts reduced the level of the mRNA of microsomal triglyceride transferase protein (MTP) in cell cultures. MTP is a protein that is necessary for the secretion of TG that is synthesized in the liver.

Based on the reports described above, the TGlowering effect of garlic could be achieved through the blockage of FAS (Qureshi *et al.*, 1983), as well as by the inhibition of MTP (Lee *et al.*, 2000).

Although the mechanism of the lipid-lowering effect of garlic on the levels of serum and hepatic TG is not clear at the time of this present study, it may be due to a combination of the inhibition of TG synthesis and secretion from the liver. A decrease in the weight of the abdominal fat pad that was concomitant with serum TG in response to 2% and 4% dietary garlic was observed at the end of the seventh week. In poultry, there is a positive correlation between the level of plasma TG and the size of the abdominal fat pad (Crespo and Steve-Garcia, 2003). On the other hand, TG that are secreted from the liver are the main source of plasma TG. Therefore, during this present study, a simultaneous reduction in the weight of the abdominal fat pad with levels of serum and liver TG may be due to the effect of garlic on suppressing hepatic TG synthesis and secretion.

In conclusion, the supplementation of the diet of broilers with garlic powder, especially at the levels of 4%, could decrease the level of TG in the liver and serum, with a slight reduction in the weight of the abdominal fat pad.

# References

- Bordia, A. (1981) Effect of garlic on blood lipids in patients with coronary heart disease. Am. J. Clin. Nutr. 34: 2100-2103.
- Carrijo, A. S., Madeira, L. A., Sartori, J. R., Pezzato, A. C., Gonclaves, J. C. (2005) Powdered garlic in the alternative feeding of two broiler chickens. Pesq. Agropec. Bras. 40:673-679
- Crespo, N., Steve-Garcia, E. (2003) Polyunsaturated fatty acids reduce insulin

and very low density lipoprotein levels in broiler chickens. Poultry Sci. 82: 1134-1139.

- Griffin, H. D., Guo, K., Windsor, D., Butterwith, S. C. (1992) Adipose tissue lipogenesis and fat deposition in line broiler chicken. J. Nutr. 122: 363-368.
- Hara, A., Radin, N. S. (1978) Lipid extraction with low toxicity solvents. Anal. Biochem. 90: 420-426.
- 6. Hermier, D. (1997) Lipoprotein metabolism and fattening in poultry. J. Nutr. 127: 805-808.
- Konjufca, V. H., Pesti, G. M., Baklli, R. I. (1997) Modulation of cholesterol levels in broiler meat by dietary garlic and copper. Poultry Sci. 76: 1264-1272.
- Leveille, G.A., Romsos, D.R., Yeh, Y., Ohea, E. K. (1975) Lipid biosynthesis in chick. A consideration of site of synthesis, Influence of diet and possible regulatory mechanism. Poultry Sci. 54: 1075-1093.
- Lin, M. C., Wang, E. J., Lee, C., Chin, K. T., Liu, D., Chiu, J. F., Kung, H. F. (2000) Garlic inhibits microsomal triglyceride transfer protein gene expression in human liver and intestinal cell lines and in rat intestine. J. Nutr. 8: 1165-1168.
- Mendez, J., Franklin, B., Gahagan, H. (1975) Simple manual procedure for determination of serum triglycerides. Clin. Chem. 21: 768-770
- 11. National Research Council. (1994) Nutrient requirement of poultry. National Academy Press. Washington, DC, USA.
- 12. Neri, P., Frings, C. S. (1973) Improved method for determination of triglyceride in serum. Clin. Chem. 19: 1201-1203.
- Pullen, D. L., Liesman, J. S., Emery, R. S. (1990) A species comparison of liver slice synthesis and secretion of triacylglycerol from nonesterified fatty acids in media. J. Anim. Sci. 68: 1395-1399.
- 14. Qureshi, A. A., Din, Z. Z., Abuirmeileh, N., Burger, W. C., Ahmad, Y., Elson, C. E. (1983a) Suppression of avian hepatic lipid



metabolism by solvent extracts of garlic: impact on serum lipids. J. Nutr. 113:1746-1755.

- 15. Qureshi, A. A., Abuirmeileh, N., Din, Z. Z., Elson, C. E., Burger, W. C. (1983b) Inhibition of cholesterol and fatty acid biosynthesis in liver enzymes and chicken hepatocytes by polar fractions of garlic. Lipids. 18: 343-348.
- 16.Saadoun, A., Leclercq, B. (1987) In vivo lipogenesis of genetically lean and fat chickens: effect of nutritional state and dietary fat. J. Nutr. 117:428-435
- 17. Yeh, Y. Y., Yeh, S. M. (1994) Garlic reduces plasma lipids by inhibiting hepatic cholesterol and triacylglycerol synthesis. Lipids.29:189-193.