

Total Antioxidant Capacity and Malondialdehyde Level in Plasma of Broiler Chicks Fed Diet Containing Different Levels of Ginger (*Zingiber officinale*)

Research Article

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ABSTRACT

This study was conducted to assess the effect of ginger supplementation on malondialdehyde (MDA) level, as an oxidative stress marker, and total antioxidant capacity (TAC) in broiler chicks. Two hundred male 1-day-old chicks were assigned randomly to four dietary treatments (basal diet as control group and basal diet containing 2.5, 5.0 and 7.5 g/kg ginger, respectively), with five replicates and ten birds per replicate. The levels of MDA and TAC were measured at days 21 and 42 of age. At day 21, mean MDA levels in chicks fed diet containing 2.5 g/kg ginger decreased numerically, and decreased significantly ($P < 0.05$) in chicks fed diet supplemented with 5 and 7.5 g/kg ginger, compared with that of control diet. At day 42, plasma MDA levels in chicks fed diets containing ginger decreased as compared with that of the control group. There were no differences for plasma MDA level among chicks fed diet containing ginger. There were significant differences ($P < 0.05$) among treatments for TAC level. At day 21, the highest mean of TAC was found in plasma of chicks fed diet containing 7.5 g/kg ginger, and the lowest one was for chicks in the control group. At this period there was no difference between TAC level of chicks fed 2.5 and 5.0 g/kg ginger. At day 42, the similar results were observed, except that ginger supplementation over 5.0 g/kg had no significant effect on TAC level. The results showed that ginger supplementation, at and over 5.0 g/kg, caused improvement in the plasma of broiler chicks, with a decrease in MDA and an increase of (TAC).

KEY WORDS broiler, ginger, malondialdehyde, plasma, total antioxidant capacity.

INTRODUCTION

Broiler chicks are constantly exposed to stress. For example, they experience vaccination, a change in ambient temperature, and many other factors that cause stress. As a result, the formation of substances (known as free radicals) increase, and these have been shown to exert undesirable effects on the tissues of the body (Roberts and Sindhu, 2009). Free radicals formed in the body are known as oxygen reactive species (ROS) and are generated in mitochondria following oxidative metabolism. When the amount of

ROS go beyond the capacity of the antioxidant capacity (enzymatic and non enzymatic), oxidative stress will occur, and malondialdehyde (MDA) as the sign of oxidative stress will increase in the blood (Ozata *et al.* 2002).

Oxidative stress causes damage to biological macromolecules (such as nucleic acids, membrane lipids and proteins) and disorders of normal metabolism and physiology (Roberts and Sindhu, 2009).

The use of ginger powder in laboratory animals has been shown to promote antioxidative properties (Kota *et al.* 2008; Mallikarjuna *et al.* 2008) and resulted in an increase

in plasma non-enzymatic antioxidant capacity in rats (Afshari *et al.* 2007). In the literature, reports on the effects of ginger powder (Afzal *et al.* 2001; Mahady *et al.* 2003) or extract (Badreldin *et al.* 2008; Mallikarjuna *et al.* 2008) on antimicrobial properties and antioxidant capacity are numerous; however, no study existing concerns the effects of the addition of ginger to diet of broiler chicks on MDA and TAC.

Therefore, the objective of the current study was to examine the effects of ginger powder on total antioxidant capacity and MDA (as a lipid peroxidation index and oxidative stress marker) in broiler chicks.

MATERIALS AND METHODS

Chickens, diets and study design

Two hundred 1-day-old broiler chicks (Ross 308) were obtained from a local hatchery. Chicks were reared in a house with separated pens (1.2×1.2 m) where the environmental conditions such as temperature, ventilation and light were controlled. Chicks were fed a starter diet from day 1 to 21 and a grower diet from day 22 to 42 (Table 1). Access to feed and water was *ad libitum* throughout the experiment. In a completely randomized design, chickens were assigned to four dietary treatments. Treatments included a control group (basal diet without ginger), and diets supplemented with 2.5, 5.0 and 7.5 g/kg ginger powder. Each treatment had five replicates (ten chicks per pen) and pens were used as replicate units.

Sample collections

On the morning of days 21 and 42 of the experiment, two birds per pen were randomly picked from each treatment group after 12 h without feed, and blood samples (5.0 mL) were taken from the wing vein into tubes containing EDTA. Blood samples were centrifuged at 1500 × g for 10 min, and the plasma samples were stored in 1.5 ml microtubes at -20 °C until analysis.

TBARS (thiobarbituric acid reactive substances) assay

MDA was determined by the TBA method, as described by Placer *et al.* (1966). Cayman's assay kit was used for this assay; 0.5 mL of plasma with 2.5 mL Trichloroacetic acid (20%) and 1 mL TBA (67%) were mixed, and then put in a hot water bath (95 °C) for 30 min. After cooling, 4 mL of butanol were added and mix and then centrifuged at 2000 × g for 10 min. After collection of the supernatant, the optical densities were measured spectrophotometrically at 532 nm.

FRAP (ferric reducing antioxidant power) assay

Total antioxidant capacity of plasma was measured according to the method of Benzie and Strain (1996). Briefly, a working solution of FRAP was provided by mixing 10 volumes of buffer acetate (300 μmol/L, pH=3.6) with 1 volume TPTZ solution in HCl (40 mM/L). After that, 1 mL solution of FeCl₃ (20 mM/L) was added and mixed. For measurement, 1.5 ml of FRAP working solution was put in the cuvette and incubated for 10 min at room temperature, then the optical density of the blank was measured spectro-

Table 1 Ingredients and chemical composition of experimental diets^a

Ingredient (%)	Starter (0 -21 day)				Grower (21-42 day)			
Ginger powder	0.00	0.25	0.50	0.75	0.00	0.25	0.50	0.75
Corn grain	50.77	50.25	49.73	49.21	55.1	54.58	54.06	53.54
Corn gluten	4.50	4.49	4.49	4.48	-	-	-	-
Soybean meal	36.75	37.03	37.13	37.23	35.8	35.90	35.99	36.09
Soybean oil	3.37	3.37	3.54	3.72	5.19	5.36	5.54	5.71
Limestone	1.33	1.33	1.33	1.33	1.07	1.07	1.07	1.07
Dicalcium phosphate	1.89	1.89	1.89	1.89	1.66	1.66	1.66	1.66
Mineral premix ^b	0.25	0.25	0.25	0.25	0.25	0.25	0.25	0.25
Vitamin premix ^c	0.25	0.25	0.25	0.25	0.25	0.25	0.25	0.25
Salt	0.30	0.30	0.30	0.30	0.25	0.25	0.25	0.25
Methionine	0.26	0.26	0.26	0.26	0.27	0.27	0.27	0.27
Lysine	0.33	0.33	0.33	0.33	0.16	0.16	0.16	0.16
Chemical composition								
ME (kcal/kg)	3025	3025	3025	3025	3150	3150	3150	3150
Crude protein (%)	24	24	24	24	22	22	22	22
Calcium (%)	1.05	1.05	1.05	1.05	0.9	0.9	0.9	0.9
Total phosphor (%)	0.75	0.75	0.75	0.75	0.69	0.69	0.69	0.69
Met + Cys (%)	1.07	1.07	1.07	1.07	0.95	0.95	0.95	0.95
Lysine (%)	1.43	1.43	1.43	1.43	1.24	1.24	1.24	1.24

^a On as-fed basis.

^b The mineral mix composition was as follows (amount in 10 g): Mg: 0.5 g; S: 0.3 g; Na: 1.0 g; Cl: 1.6 g; Cu: 6.0 mg; I: 0.2 mg; Fe: 45.0 mg; Mn: 59 mg; Se: 0.2 mg and Zn: 29 mg.

^c The vitamin mix composition was as follows (amount in 10 g): vitamin A palmitate: 4000 IU; cholecalciferol: 1000 IU; vitamin E acetate: 50 IU; Menadione sodium bisulfite: 0.5 mg; Biotin: 0.2 mg; Cyanocobalamin: 10 μg; Folic acid: 2 mg; Nicotinic acid: 30 mg; Calcium pantothenate: 16 mg; Pyridoxine-HCl: 7 mg; Riboflavin: 6 mg; Thiamin-HCl: 6 mg.

photometrically at 532 nm. For test samples, 50 μ L of plasma replaced by the working solution, and change in absorbance was measured.

Statistical analysis

The Kolmogorov-Smirnov test was used to test the normal distribution of the data before statistical analysis was performed.

Statistical analyses were conducted with the general linear model procedure of SAS for Windows version 9.1 (SAS, 1998) to determine if variables differed between groups. Whenever significant differences were found, mean values were compared by the Tukey test. A probability value of less than 0.05 was considered significant.

RESULTS AND DISCUSSION

Results of plasma MDA level at days 21 and 42 of age are shown in Figure 1 and 2. At 21 days of age, mean MDA level in chicks fed diet containing 2.5 g/kg ginger decreased numerically, but in chicks fed diets supplemented with 5.0 and 7.5 g/kg ginger decreased significantly ($P<0.05$), compared to chicks fed the control diet. At day 42, plasma MDA levels in chicks fed the diet containing ginger decreased, as compared with that of the control group. There were no differences for plasma MDA level among chicks fed diets containing ginger.

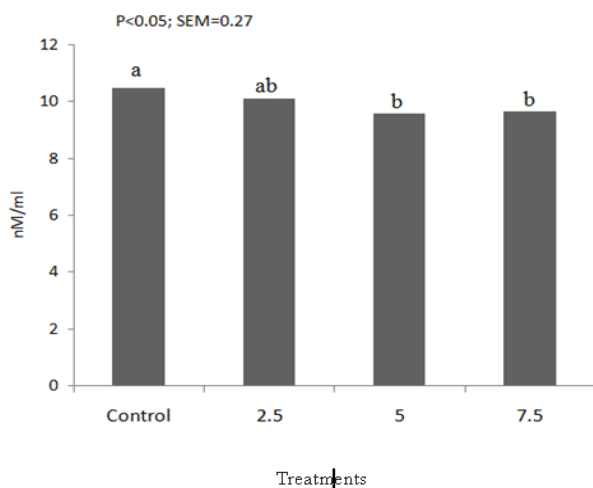


Figure 1 Oxidative stress index (MDA, as μ mol/mL) in plasma of broiler chicks at day 21 of age

Mean plasma TAC levels at day 21 and 42 of age are shown in Figure 3 and 4. There were differences ($P<0.05$) among treatments for TAC level. At day 21, the highest mean of TAC was found in plasma of chicks fed diet containing 7.5 g/kg ginger, and the lowest one was for chicks fed the control diet. At this period, there was no difference between TAC level of chicks fed 5 and 7.5 g/kg ginger.

At day 42, similar results were observed, except that ginger supplementation over 5.0 g/kg had no significant effect on TAC level.

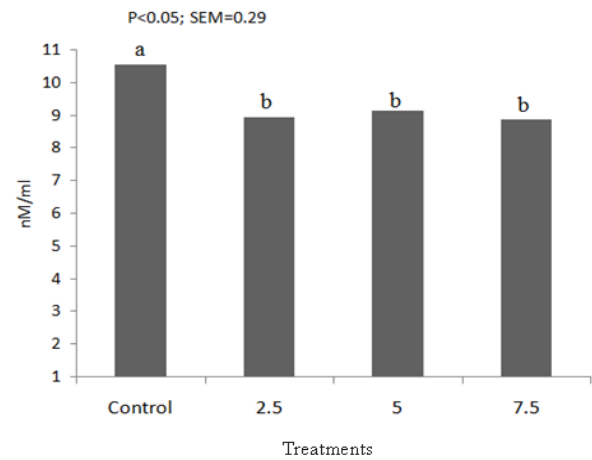


Figure 2 Oxidative stress index (MDA, as μ mol/mL) in plasma of broiler chicks at day 42 of age

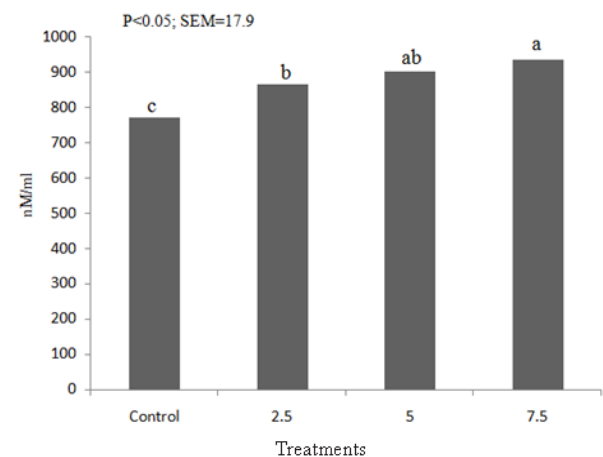


Figure 3 Total antioxidant capacity of plasma (TAC, as μ mol per ml) in broiler chicks at day 21 of age

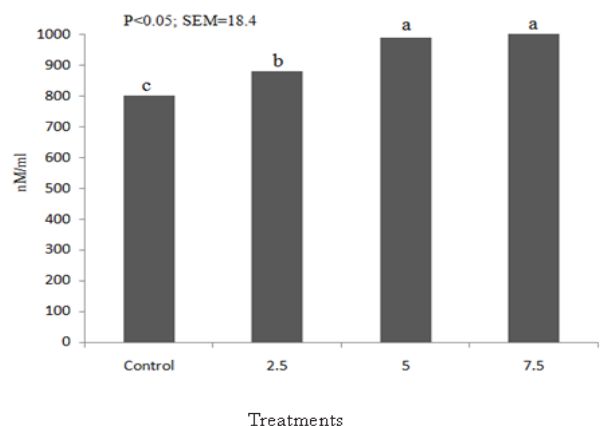


Figure 4 Total antioxidant capacity of plasma (TAC, as μ mol per ml) in broiler chicks at day 42 of age

Rapid growth rate and various challenges in broiler chicks cause increasing free radicals, which result in increased oxidative stress. The birds become susceptible to many disorders, such as ascites and sudden death syndrome (Roberts and Sindhu, 2009). Consumption of products rich in antioxidants as additives in broiler diets could increase physiological antioxidant defenses, which results in a decrease in oxidative stress. Therefore, the present study investigated the effect of ginger powder supplementation on MDA as one of the blood oxidative stress indicators of plasma non-enzymatic antioxidant capacity in broiler chicks. The MDA is formed as an end product of lipid peroxidation, and the birds fed diets containing ginger powder had lower MDA than the control group. This finding indicates that lipid peroxidation was reduced by ginger powder via enhancing antioxidative action, which is in agreement with the results of Zhang *et al.* (2009). They reported that MDA concentration in the plasma was reduced by ginger supplementation and attributed to the increase of antioxidant enzymatic activity. Our finding is also in agreement with the reports of Bayraktar *et al.* (2011) and Erdogan *et al.* (2005). They found that usage of an antioxidant additive in broiler diets could prevent the oxidative stress and decrease MDA as lipid peroxidation marker. The level of TAC in plasma of chicks fed diets containing ginger powder increased. This result indicates that ginger powder had antioxidant properties. Ginger powder contains many of compounds that have biological activities, including antioxidants (Nakatani, 2000; Rababah *et al.* 2004) and various pharmacological effects (Chrubasik *et al.* 2005; Ali *et al.* 2008). The potential active constituents in ginger are the gingerols, shogaols, gingerdiol, gingerdione, and some related phenolic ketone derivatives (Kikuzaki and Nakatani, 1996; Fuhrman *et al.* 2000), which act as antioxidants. Our results are in agreement with Krishnakantha and Lokesh (1993), who reported that ginger extract, has antioxidative properties and scavenges superoxide anion and hydroxyl radicals. Afshari *et al.* (2007) also observed that plasma antioxidant capacity increased after consumption of ginger in Wistar rats and elevation of antioxidant capacity raised the antioxidant scavenging capacity of the body and attenuated free radical-induced damage.

CONCLUSION

The results of this study showed that, ginger supplementation, at and over 5.0 g/kg, caused improvement in plasma of broiler chicks by decreasing MDA and increasing TAC.

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REFERENCES

- Afshari A.T., Shirpoor A., Farshid A., Saadatian R., Rasmi Y., Saboory E., Ilkhanizadeh B. and Zillameh A. (2007). The effect of ginger on diabetic nephropathy, plasma antioxidant capacity and lipid peroxidation in rats. *Food Chem.* **101**, 148-153.
- Afzal M., Al-Hadidi D., Menon M., Pesek J. and Dhami M.S. (2001). Ginger: an ethno medical, chemical and pharmacological review. *Drug Metabol. Drug Interact.* **18**, 159-190.
- Ali B.H., Blunden G., Tanira M.O. and Nemmar A. (2008). Some phytochemical, pharmacological and toxicological properties of ginger (*Zingiber officinale roscoe*): a review of recent research. *Food Chem. Toxicol.* **46**, 409-420.
- Badreldin, H.A., Gerald, B., Musbah, O. and Nemmar A. (2008) Some phytochemical, pharmacological and toxicological properties of ginger (*Zingiber officinale Roscoe*): a review of recent research. *Food Chem. Toxicol.* **46**, 409-420.
- Bayraktar H., Altan Ö., Açıkgöz Z., Baysal S.H. and Şeremet C. (2011). Effects of oxidized oil and vitamin E on performance and some blood traits of heat-stressed male broilers. *South African J. Anim. Sci.* **41**, 321-329.
- Benzie I.F. and Strain J.J. (1996). The ferric reducing ability of plasma (FRAP) as a measure of "antioxidant power: the FRAP assay. *Anal. Biochem.* **239**, 70-76.
- Chrubasik S., Pittler M.H. and Roufogalis B.D. (2005). A comprehensive review on the ginger effect and efficacy profiles. *Phytomed.* **12**, 684-701.
- Erdogan Z., Erdogan S., Aksu T. and Baytok E. (2005). The effects of dietary lead exposure and ascorbic acid on performance, lipid peroxidation status and biochemical parameters of broilers. *Turk J. Vet. Anim. Sci.* **29**, 1053-1059.
- Fuhrman B., Rosenblat M., Hayek T., Coleman R. and Aviram M. (2000). Ginger extract consumption reduces plasma cholesterol, inhibits LDL oxidation and attenuates development of atherosclerosis in atherosclerotic, apo lipoprotein E-deficient mice. *J. Nutr.* **130**, 1124-1131.
- Kikuzaki H. and Nakatani N. (1996). Cyclic diarylheptanoids from rhizomes of *Zingiber officinale*. *Phytochem.* **43**, 273-277.
- Kota N., Krishna P. and Polasa K. (2008). Alterations in antioxidant status of rats following intake of ginger through diet. *Food Chem.* **106**, 991-996.
- Krishnakantha T.P. and Lokesh B.R. (1993). Scavenging of superoxide anions by spice principles. *Ind. J. Biochem. Biophys.* **30**, 133-134.
- Mahady G.B., Pendland S.L., Yun G.S., Lu Z.Z. and Stoia A. (2003). Ginger (*Zingiber officinale Roscoe*) and the gingerols inhibit the growth of Cag A+ strains of *Helicobacter pylori*. *Anticancer. Res.* **23**, 3699-3702.

- Mallikarjuna K., Chetan P.S., Reddy K.S. and Rajendra W. (2008). Ethanol toxicity: rehabilitation of hepatic antioxidant defense system with dietary ginger. *Fitoterapia*. **79**, 174-178.
- Ozata M., Mergen M., Oktenli C., Aydin A., Sanisoglu S.Y., Bolu E., Yilmaz M.I., Sayal A., Isimer A. and Ozdemir I.C. (2002). Increased oxidative stress and hypozincemia in male obesity. *Clin. Biochem.* **35**, 627-631.
- Placer Z.A., Cushman L.L. and Johnson B.C. (1966). Estimation of product of lipid peroxidation (malondialdehyde) in biochemical systems. *Anal. Biochem.* **16**, 359-364.
- Rababah T.M., Hettiarachchy N.S. and Horax R. (2004). Total phenolics and antioxidant activities of fenugreek, green tea, black tea, grape seed, ginger, rosemary, gotu kola, and ginkgo extracts, vitamin E, and tert-butylhydroquinone. *J. Agric. Food Chem.* **52**, 5183-5186.
- Roberts C.K. and Sindhu K.K. (2009). Oxidative stress and metabolic syndrome. *Life. Sci.* **84**, 705-712.
- SAS Institute. (1998). SAS User's Guide: Statistics. SAS Institute Inc. USA.
- Zhang G.F., Yang Z.B., Wang Y., Yang W.R., Jiang S.Z. and Gai G.S. (2009). Effects of ginger root (*Zingiber officinale*) processed to different particle sizes on growth performance, antioxidant status, and serum metabolites of broiler chickens. *Poult. Sci.* **88**, 2159-2166.
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