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RESEARCH ARTICLE

# Antioxidant Activity of Cinnamon Bark Oil (*Cinnamomum zeylanicum* L.) in Japanese Quails Under Thermo Neutral and Heat Stressed Conditions

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## Summary

The essential oil from bark of *Cinnamomum zeylanicum Lauraceae* was analyzed by GC and GC-MS systems in this research. The essential oil was obtained by hydrodistillation, in 0.7 (v/w) oil yields. Twelve constituents representing 99.2% of the cinnamon oil were identified. The major compounds in the oil were *cinnamaldehyde* (88.2%), *benzyl alcohol* (8.0%) and *eugenol* (1.0%). To determine antioxidant activity of the cinnamon oil, a total of 180 quails, fifteen-days-old, were allocated into 6 groups consisting of 10 birds of 3 replicates according to balanced gender and inital live weight. The birds were kept in wire cages in a temperature controlled room at 22°C for 24 h/d in thermo-neutral (TN) groups. For heat-stress (HS) groups, the birds were exposed to 34°C for 8 h/d (from 9:00 to 17:00), and later 22°C 16 h/d was performed. Relative humidity was approximately 60-65%. Basal diet was given to control groups in both TN and HS. The birds were fed with the basal diet supplemented 250 or 500 ppm cinnamon oil in the other experimental groups. Heat stress increased the malondialdehyde (MDA) levels of liver ($P<0.001$), heart ($P<0.01$) and kidney ($P<0.05$). It also induced superoxide dismutase (SOD) production of liver ($P<0.001$) and kidney ($P<0.05$). Glutathione peroxidase (GSH-Px) activity and glutathione (GSH) level of liver ($P<0.001$, $P<0.05$) and heart ($P<0.001$, $P<0.05$) were found lower under HS condition. Cinnamon oil supplementation to diet significantly increased antioxidant enzyme activity and GSH level of the tissues in both enviromental conditions ($P<0.01$). Dose of 500 ppm cinnamon oil had strong effect on antioxidant activity of the internal organs ($P<0.01$). In conclusion, cinnamon oil supplementation to diet reduced the adverse effects of heat stress and resulted the protective effect on the internal organs by activating antioxidant mechanism.

***Keywords:*** *GC-MS, Cinnamon oil, Lipid peroxidation, Antioxidant activity, Quail, Heat stress*

# Termo-Nötral ve Sıcaklık Stresi Koşullarındaki Japon Bıldırcınlarında Tarçın Kabuğu Yağının (*Cinnamomum zeylanicum* L.) Antioksidan Aktivitesi

## Özet

Bu araştırmada, tarçın (*Cinnamomum zeylanicum Lauraceae*) kabuğundan elde edilen uçucu yağ GC and GC-MS sistemleri ile analiz edilmiştir. Uçucu yağın elde edilmesinde hidrodistilayon 0.7 (v/w) metodu kullanılmıştır. Elde edilen yağın %99. 2 sini oluşturan oniki bileşen tanımlanmıştır. Tarçın yağının önemli bileşenlerini *sinamaldehid* (%88.2), *benzil alkol* (%8.0) ve *öjenol* (%1.0) oluşturmaktadır. Tarçın yağının antioksidan aktivitesini belirlemek için toplam 180 adet on beş günlük bıldırcın, altı alt gruba ayrılmıştır. Her grup on bıldırcın içeren üç tekerrürden oluşturulmuş, gruplar başlangıç canlı ağırlığı ve cinsiyet bakımından dengelenmiştir. Bıldırcınlar sıcaklık kontürollü odalarda tel kafeslerde, termo-nötral (TN) grupta 22°C'de 24 saat/gün şeklinde barındırılmışlardır. Sıcaklık stresi (HS) grupları günde 8 saat süresince (9:00'dan 17:00'ye/tüm araştırma) 34°C'ye ve daha sonra 22°C'ye 16 saat/gün sıcaklığa maruz bırakılmışlardır. Bağıl nem yaklaşık %60-65 dir. TN ve HS şartlarında kontrol grubundaki bıldırcınlara temel yem verilmiştir. Diğer deneme grupları temel yeme 250 ve 500 ppm tarçın yağı ilave edilen yemle beslenmişlerdir. Sıcaklık stresi karaciğer ($P<0.001$), kalp ($P<0.01$) ve böbrek ($P<0.05$) dokularında malondialdehit (MDA) düzeyini yükseltmiştir. Stres karaciğer ($P<0.001$) ve böbrek ($P<0.05$) dokularında süperoksit dismutaz (SOD) aktivitesini artırırken, karaciğer ($P<0.001$, $P<0.05$) ve kalp dokularında ($P<0.001$, $P<0.05$) sırasıyla glutatyon peroksidaz (GSH-Px) aktivitesi ile glutatyon (GSH) seviyesini azaltmıştır. Yeme ilave edilen tarçın yağı her iki çevre şartında dokuların antioksidan enzim aktiviteleri ile GSH düzeyini önemli ölçüde yükseltmiştir ($P<0.01$). Tarçın yağının 500 ppm düzeyindeki dozu güçlü antioksidan özellik göstermiştir ($P<0.01$). Sonuç olarak, yeme katılan tarçın yağı sıcaklık stresinin olumsuz etkilerini azaltmış ve antioksidan metabolizmayı aktive ederek iç organlar üzerinde koruyucu etki göstermiştir.

***Anahtar sözcükler:*** *GC-MS, Tarçın yağı, Oksidatif stres, Antioksidan aktivite, Bıldırcın, Sıcaklık stresi*

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# INTRODUCTION

Heat stress (HS) is a very serious situation for poultry and can be described as difficulty achieving a balance between body heat production and heat loss. Air temperature, humidity, radiant heat and air speed are important factors causing acute or chronic HS in poultry. Acute HS refers to short and sudden periods of the extremely conditons, whereas chronic HS refers to extended periods of elevated the conditions [1,2]. Under the long term stress condition or repeated stress, birds start to fatigue and weak [2,3]. Long-term regulation of body in chronic stress conditions are characterized by adrenal cortical hypertrophy and increased synthesis and release of adrenal glucocorticoids, known as corticosterone (CORT) in bird [4,5]. Administration of CORT may initially induce the formation of reactive oxygen species (ROS) as indirectly reflected by an increase in lipid peroxidation (LP) [6,7]. The ROS including hydrogen peroxide ($H_2O_2$), hypochlorous acid (HClO) and free radicals such as the hydroxyl radical (·OH) and the superoxide anion ($O_2^-$) are produced in cells. ROS is a natural by-product of the routine metabolism of oxygen, however under the environmental stress (e.g., heat stress), levels of ROS can increase dramatically. This may resulted with significant damage in the cell structure. Cumulatively, this is known as oxidative stress [6]. Malondialdehyde (MDA) is one of the final products of polyunsaturated fatty acids peroxidation in the cells. An increase in ROS causes overproduction of MDA, it is a reactive aldehyde and is one of the many reactive electrophile species that cause toxic stress for the cells [8]. Superoxide dismutase (SOD) is a group of metalloenzymes whose function appears to be protection of cells from the toxic effects of the endogenously generated superoxide radicals [9]. Glutathione (GSH) is an antioxidant that play a major role in a redox potential regulation, detoxication of ROS, and also serves as a store and transport form of cysteine as well [10]. Glutathione peroxidase enzyme (GSH-Px) provides a mechanism for detoxification of peroxides (e.g. $H_2O_2$) and a wide variety of organic peroxides (R-OOH) to alcohols (R-OH) and water using cellular glutathione in living cells [9]. External antioxidants can directly react with free oxygen radicals, genotoxic susbtances or carcinogenics, by chelating complexes with transition metals, act as reducing agents, induce the production of antioxidative enzymes [11,12].

Cinnamon *(Cinnamomum zeylanicum* L.) is native to tropical Asia, especially Sri Lanka and India. Cinnamon bark is used as spices and for the production of essential oil. It has lots of medical properties [13] and antioxidant activity [14]. Plant tissue of cinnamon has a wide variety of phenolic compounds such as flavonoids, isoflavones, flavones, catechin and other phenolics. The phenolic compounds are dominant antioxidants that exhibit scavenging efficiency on ROS. The plant tissue has also strong antioxidant capacity because of richness from some antioxidant vitamins and minerals [15,16].

The objective of the present study is to determine antioxidant property of *Cinnamomum zeylanicum* bark oil in Japanese quails reared under thermo neutral and heat stressed condition.

# MATERIAL and METHODS

### *Experimental Design*

A total of 180 Japanese (*Coturnix coturnix Japonica*) quails, fifteen-days-old, obtained from a commercial company were used for the experiment. This study was undertaken after ethical approval of Firat University (Official form date and number: 20.01.2011 and 2011/15). The experiment was conducted at the Poultry Unit of Firat University. The birds were assigned to experimental groups at the beginning of the study with a balanced gender and inital live weight. The experimental design were performed according to 2 (thermo neutral-TN, high ambient temperature-HS) × 3 (cinnamon oil levels: 0, 250, 500 ppm) factorial design, 6 treatment groups consisting of 10 birds with 3 replicates. The birds were kept in wire cages in a temperature controlled room at 22°C for 24 h/d in TN groups. For HS groups, the birds were exposed to 34°C for 8 h/d (from 9:00 to 17:00), and later 22°C 16 h/d was performed. Relative humidity was approximately 60-65%. Basal diet was given to control groups of both TN and HS. The birds were fed with the basal diet supplemented 250 or 500 ppm cinnamon oil in the other experimental groups. The cinnamon oil was mixed in a carrier (zeolite), then added to the basal diet at a level of 1 kg/ton. The concentration of the volatile components in cinnamon oil was shown at *Figure 1* (shown as percentage peak areas of GC-MS). Diets and fresh water were given *ad libitum*. Light was provided continuously (24 h) throughout the experiment. Ingredients and chemical composition of the basal diet were shown at *Table 1*.

At the end of the study (43rd day) six male and six female quails from each experimental group whose body weight near the group average were slaughtered. After slaughtering process, liver, heart and kidney tissues were obtained from the carcasses and wrapped with aluminum foil and stored at -20°C until analysis.

### *Chemical Analysis*

Chemical composition of feed ingredients (dry matter, crude protein, ash and ether extract) were analyzed according to the AOAC [17] procedures and crude fibre was determined by the method of CRAMPTON and MAYNARD [18].

### *Gas Chromatographic (GC) Analysis*

The essential oil was analyzed using HP 6890 GC equipped with and FID detector and an HP- 5 MS (30 m x 0.25 mm i.d., film tickness 0.25 µm) capillary column was used. The column and analysis conditions were the same as in GC-MS. The percentage composition of the essential oil

**Table 1.** *Ingredients and chemical composition of basal mix diet*
**Tablo 1.** *Bazal karma yemin bileşimi ve kimyasal kompozisyonu*

| Feed Ingredients | % | Nutritional Composition | % |
|---|---|---|---|
| Maize | 29.03 | Dry matter | 88.25 |
| Wheat | 25.00 | Crude protein | 23.87 |
| Soybean meal (48 CP) | 34.29 | Crude fibre | 2.55 |
| Corn Gluten | 4.10 | Ether extract | 4.75 |
| Vegetable oil | 2.92 | Ash | 5.45 |
| Dicalcium phosphate | 2.02 | Calcium **** | 1.00 |
| Ground limestone | 0.87 | Total phosphorus**** | 0.79 |
| $NaHCO_3$ | 0.12 | ME, kcal/kg**** | 2897 |
| Salt | 0.28 | | |
| DL-Metiyonin | 0.02 | | |
| Vitamin mix * | 0.25 | | |
| Mineral mix** | 0.10 | | |
| Additive*** | 1.00 | | |

** Vitamin premix supplied per 2.5 kg; Vitamin A 12.000.000 IU; vitamin $D_3$ 2.000.000 IU; vitamin E 35.000 mg; vitamin $K_3$ 4.000 mg; vitamin $B_1$ 3.000 mg; vitamin $B_2$ 7.000 mg; Niacine 20.000 mg; Calcium D-pantotenat 10.000 mg; vitamin $B_6$ 5.000 mg; vitamin $B_{12}$ 15 mg; Folik Asit 1.000 mg; D-Biotin 45 mg; vitamin C 50.000 mg; Choline chloride 125.000 mg; Canthaxanthin 2.500 mg; Apo Karotenoik Acid Ester 500 mg , ** Mineral premix supplied per kg; Mn 80.000 mg; Fe 60.000 mg; Zn 60.000 mg; Cu 5.000 mg; Co 200 mg; I 1.000 mg; Se 150 mg, *** Group Cinnamon 0 (1000 g zeolit); Group Cinnamon 250 (25 g cinnamon oil+975 g zeolit); Group Cinnamon 500 (50 g cinnamon oil + 950 g zeolit), **** Calculated*

was computed from GC - FID peak areas without correction factors.

### *Gas Chromatography/Mass Spectrometry (GC-MS) Analysis*

The oils were analyzed by GC-MS, using a Hewlett Packard system. HP- Agilent 5973 N GC-MS system with 6890 GC in Plant Products and Biotechnology Res. Lab. in Firat University. HP-5 MS column (30 m x 0.25 mm i.d., film tickness 0.25 µm) was used with helium as the carrier gas. Injector temperature was 250°C, split flow was 1 ml/min. The GC oven temperature was kept at 70°C for 2 min and programmed to 150°C at a rate of 10°C/min and then kept constant at 150°C for 15 min to 240°C at a rate of 5°C/min. Alkanes were used as reference points in the calculation of relative retention indices (RRI). MS were taken at 70 eV and a mass range of 35-425. Component identification was carried out using spectrometric electronic libraries (Wiley, Nist).

### *Lipid Peroxidation*

The malondialdehyde (MDA) level was measured in serum using the thiobarbituric acid reaction described by PLACER et al.[19]. The quantification of thiobarbituric acid reactive substances was determined by comparing the absorption to the standard curve of MDA equivalents generated by acid catalyzed hydrolysis of 1,1,3,3 tetra-methoxypropane. Each sample was assayed in duplicate, and the assay coefficients of variation for MDA were less than 3%.

### *Superoxide Dismutase (SOD)*

The plasma SOD activity was measured using xanthine and xanthine oxidases to generate superoxide radicals which react with nitroblue tetrazolium (NBT) [20]. Briefly, each sample was diluted 1:10 with phosphate buffer (50 mM, pH 7.5). The assay solution containing sodium-carbonate buffer (50 mM, pH 10), 0.1 mM xanthine, 0.025 mM NBT, 0.1 mM EDTA, xanthine oxidase (0.1 U/mL in ammonium sulfate 2 M) and sample were mixed in a cuvette. One unit of SOD activity was defined as the amount of enzyme required to cause inhibition of NBT. SOD activity was then measured at 560 nm by the degree of inhibition of this reaction on a spectrophotometer and expressed as U/mL.

### *Glutathione Peroxidase (GSH-Px)*

The GSH-Px activity was determined according to LAWRENCE and BURK [21]. The reaction mixture consisted of 50 mM potassium phosphate buffer (pH 7.0), 1 mM ethylene diamine tetra acetic acid (EDTA), 1 mM sodium azide (NaN3), 0.2 mM reduced nicotinamide adenine dinucleotide phosphate (NADPH), 1 IU/ml oxidized glutathione (GSSG)-reductase, 1 mM GSH, and 0.25 mM hydrogen peroxide ($H_2O_2$). Enzyme source (0.1 ml) was added to 0.8 ml of the above mixture and incubated at 25°C for 5 min before initiation of the reaction with the addition of 0.1 ml of peroxide solution. The absorbance at 340 nm was recorded for 5 min on a spectrophotometer. The activity was calculated from the slope of the lines as micromoles of NADPH oxidized per minute. The blank value (the enzyme was replaced with distilled water) was subtracted from each value.

### *Reduced Glutathione (GSH)*

The GSH content of the serum was measured at 412 nm by the method of SEDLAK and LINDSAY [22]. The samples were precipitated with 50% trichloroacetic acid and then centrifuged at 1000 × g for 5 min. The reaction mixture contained 0.5 ml of supernatant, 2.0 ml of Tris-EDTA buffer (0.2 M; pH 8.9) and 0.1 ml of 0.01 M 5,5'-dithio-bis-2-nitrobenzoic acid. The solution was kept at room temperature for 5 min, and then read at 412 nm on the spectrophotometer.

### *Statistical Analysis*

Data were evaluated by using GLM (General Linear Model) procedure (2x3 factorial design), significant differences were further subjected to Duncan's multiple range test (SPSS) [23]. The results were considered significant when P values were lower than 0.05.

## RESULTS

Twelve compounds were identified by GS-MS in C. *zeylanicum* bark oil representing 99.2% of the total oil (*Fig. 1*

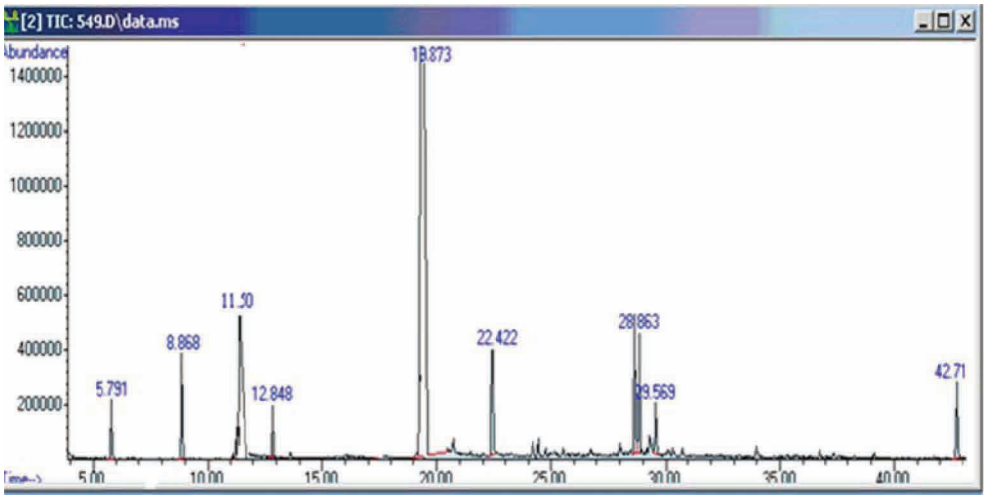


**Fig 1.** Total ion chromatogram of Cinnamon (*Cinnamomum zeylanicum L.)* bark volatile oil.

**Şekil 1.** Tarçın (*Cinnamomum zeylanicum L.*) kabuğu uçucu yağının toplam iyon kromotogramı

***Table 2.*** *Chemical composition of cinnamon (Cinnamomum zeylanicum L.) bark volatile oil*

***Tablo 2.*** *Tarçın (Cinnamomum zeylanicum L.) kabuğu uçucu yağının kimyasal kompozisyonu*

| Peak No | RT (min) | RRI | Compounds | Peak Area (%) | Identification Method |
|---|---|---|---|---|---|
| 1 | 5.79 | 976 | p-Xylene | 0.2 | GC, GC-MS |
| 2 | 8.86 | 1044 | Benzaldehyde | 0.3 | |
| 3 | 11.50 | 1102 | Benzyl Alcohol | 8.1 | |
| 4 | 12.84 | 1131 | Formic acid | 0.2 | |
| 5 | 13.62 | 1148 | α-Terpinolene | 0.1 | |
| 6 | 15.63 | 1192 | Benzenepropanol | 0.1 | |
| 7 | 16.69 | 1216 | α-Terpineol | 0.1 | |
| 8 | 19.87 | 1286 | Cinnamaldehyde | 88.2 | |
| 9 | 22.41 | 1341 | Eugenol | 1.0 | |
| 10 | 28.86 | 1483 | Cinnamaldehyde propylene glycol acetal | 0.5 | |
| 11 | 29.56 | 1493 | 1H-Cycloprop[e] azulene | 0.1 | |
| 12 | 42.71 | 1788 | Benzyl cinnamate | 0.3 | |
| Total | | | | 99.2 | |

***RT:*** *Retention Time,* ***RRI:*** *Relative Retention Indices,* ***RRI=*** *((RT*100)+3865)/4.55*

and *Table 2*). Three of them are major compounds including *cinnamaldehyde* (88.2%), *benzyl alcohol* (8.0%) and *eugenol* (1.0%). Other nine compounds are minor containing in amounts less than 1%, including *cinnamaldehyde propylene glycol acetal* (0.5%), *benzaldehyde* (0.3%), *benzyl cinnamate* (0.3%), *p-xylene* (0.2%) *formic acid* (0.2%), *α-terpinolene* (0.1%), *benzenepropanol* (0.1%), *α-terpineol* (0.1%), *1H-cycloprop[e]azulene* (0.1%).

Heat stress increased the MDA levels of liver (P<0.001), heart (P<0.01) and kidney (P<0.05) tissues *(Table 3)*. SOD production of liver (P<0.001) and kidney (P<0.05) were found higher, however GSH-Px activity and GSH production of liver (P<0.001, P<0.05) and heart (P<0.001, P<0.05) were found lower under the HS condition. Cinnamon oil addition to diet decreased MDA levels of liver (P<0.05), heart (P<0.001) and kidney (P<0.05) tissues under HS condition, the current dose of cinnamon oil did not have significant effect in that condition. MDA level of heart was decreased (P<0.001) with the dose of cinnamon oil in TN condition even 500 ppm level had better effect. SOD activity of liver (P<0.001) and heart (P<0.05) increased in cinnamon oil groups in both enviromental conditions. GSH-Px activity of heart (P<0.05) in thermo-neutral condition and GSH level of heart (P<0.01) and kidney (P<0.01) were found to be higher in cinnamon groups in the both conditions.

## DISCUSSION

The results of the current study show that twelve compounds representing 99.2% of the total essential oil of C. *zeylanicum* bark were identified. The major compounds in the essential oil were *cinnamaldehyde* (88.2%), *benzyl alcohol* (8.0%) and *eugenol* (1.0%). Other components analyzed in the oil were less than 1%. The composition of the essential oil of *C. zeylanicum* is quite variable, depending on the locality of growth and different part of the plant. However, *eugenol* is main component of the oil from leaf and *cinnamaldehyde* for the oil from bark [24,25]. WANG et al.[26] reported that twenty one volatile compounds identified from the essential oil of *C. zeylanicum* leaf, including aldehydes, alcohols, alkanes, ketones, ethers and sulfides. *Eugenol* (79.75%) was the major volatile component instead of *trans-cinnamaldehyde* (16.25%). FICHI et al.[27] stressed that *eugenol* (76.1%), *caryophyllene* (6.7%) and *linalool* (3.7%) were the major components in the chemical composition of the essential oil of *C. zeylanicum* leaves. Similar to the current study, UNLU et al.[28] analyzed the essential oil from the bark of *C. zeylanicum* by using GC-MS. Nine constituents representing 99.24% of the total oil were identified. The major compounds in the oil were *cinnamaldehyde* (68.95%), *benzaldehyde* (9.94%) and *cinnamyl acetate* (7.44%). YANG et al.[29] showed that *C. zeylanicum* bark essential oil was composed of three major [*cinnamaldehyde* (58.1%), *benzaldehyde* (12.2%) and

***Table 3.*** *Malondialdehyde (MDA), superoxide dismutase (SOD), glutathione peroxidase (GSH-Px) and glutathione (GSH) levels of inner organs in experimental groups*

***Tablo 3.*** *Deneme gruplarında iç organların malondialdehit (MDA), süperoksit dismutaz (SOD), glutatyon peroksidaz (GSH-Px) ve glutatyon (GSH) düzeyleri*

| Traits | HS-Heat Stress | | | TN- Thermo Neutral | | | SEM | P- Statistical Significance | | |
|---|---|---|---|---|---|---|---|---|---|---|
| | Cinnamon Oil, ppm | | | Cinnamon Oil, ppm | | | | Main Effects | | CO*EC |
| | 0 | 250 | 500 | 0 | 250 | 500 | | Environmental Conditions (EC) | Dose of Cinnamon Oil (CO) | |
| **MDA (nmol/mL)** | | | | | | | | | | |
| Liver | 12.29$^{a}$ | 8.21$^{b}$ | 8.25$^{b}$ | 6.13$^{A}$ | 5.89$^{A}$ | 5.74$^{A}$ | 0.46 | *** | * | NS |
| Heart | 191.33$^{a}$ | 129.53$^{b}$ | 129.93$^{b}$ | 131.49$^{A}$ | 123.32$^{B}$ | 97.96$^{C}$ | 5.39 | ** | *** | NS |
| Kidney | 62.82$^{a}$ | 51.95$^{b}$ | 47.92$^{b}$ | 51.43$^{A}$ | 45.50$^{A}$ | 46.26$^{A}$ | 1.59 | * | * | NS |
| **SOD (U/Hb/mL)** | | | | | | | | | | |
| Liver | 1.24$^{b}$ | 2.06$^{ab}$ | 2.37$^{a}$ | 1.28$^{B}$ | 1.35$^{AB}$ | 1.58$^{A}$ | 0.08 | *** | *** | ** |
| Heart | 6.60$^{b}$ | 8.26$^{a}$ | 7.63$^{ab}$ | 6.73$^{B}$ | 8.20$^{A}$ | 8.15$^{A}$ | 0.21 | NS | ** | NS |
| Kidney | 2.92 | 3.17 | 3.34 | 2.77 | 2.95 | 2.64 | 0.08 | * | NS | NS |
| **GSH-Px (U/g Hb)** | | | | | | | | | | |
| Liver | 0.05 | 0.06 | 0.06 | 0.07 | 0.08 | 0.06 | 0.00 | *** | NS | NS |
| Heart | 0.29$^{a}$ | 0.32$^{a}$ | 0.29$^{a}$ | 0.35$^{B}$ | 0.33$^{B}$ | 0.44$^{A}$ | 0.01 | *** | * | NS |
| Kidney | 0.10 | 0.11 | 0.09 | 0.10 | 0.11 | 0.10 | 0.00 | NS | NS | NS |
| **GSH (nmol/mL)** | | | | | | | | | | |
| Liver | 0.011 | 0.010 | 0.010 | 0.012 | 0.012 | 0.015 | 0.00 | * | NS | NS |
| Heart | 0.030$^{b}$ | 0.040$^{a}$ | 0.029$^{b}$ | 0.025$^{B}$ | 0.034$^{A}$ | 0.030$^{AB}$ | 0.00 | * | ** | NS |
| Kidney | 0.018$^{b}$ | 0.013$^{b}$ | 0.024$^{a}$ | 0.020$^{B}$ | 0.016$^{B}$ | 0.036$^{A}$ | 0.00 | NS | ** | NS |

***CO*EC:*** *Interaction between main effects, SEM: Standart Error of Mean,* ***NS:*** *P>0.05, * P<0.05, **P<0.01, ***P<0.001,* $^{a,b,c \text{ and } A,B,C}$***:*** *Meas with different superscripts in a sub group significantly differ*

*eugenol* (5.1%)] and six minor constituents by GC-MS. The essential oil of cinnamon bark is known to be a unique aromatic monoterpene-rich natural source, with *trans-cinnamaldehyde* (45.62%) as the major constituents. It contains relatively high amounts of phenolic compounds (18.2% of the oil), their phenolic group plays an important role in antioxidant activity, which act as hydrogen donor [30,31]. VARALAKSHMI et al.[32] reported that bark of *C. zeylanicum* was a potential source of natural antioxidants, and could be used in any preparations for combating free radical mediated damage to the body. MATHEW and ABRAHAM [33] showed that cinnamon exracts contain a number of anti-oxidant compounds which could effectively scavenge reactive oxygen species including superoxide anions and hydroxyl radicals as well as other free radicals. CIFTCI et al.[34] suggested that cinnamon oil might play an important role as an endogenous antioxidant metabolism and could also be applicable as a protective agent against tissue damage. Increasing MDA levels of liver, heart and kidney in the current study might be due to the heat stress condition. The liver tissue was quite affected from HS force compared to heart and kidney. HS induced SOD production in liver and kidney. It decreased the GSH-Px and GSH production of liver and heart. Similarly, YANG et al.[35] mentioned about heat stress induced a significant production of ROS, function of the mitochondrial respiratory chain, anti-oxidative enzymes such as SOD, CAT, GSH-Px activity and formation of MDA. Supplementation of cinnamon oil of both doses to diet reduced MDA production in liver, heart and kidney, especially under HS condition. SCHMIDT et al.[36] mentioned about among all oxygen radicals the hydroxyl radical (OH•) was the most reactive and damages diverse biomolecules, and cinnamon oil showed high hydroxyl radical-scavenging activity because of its phenolic structure. FAIX et al.[37] observed that 0.1% level *C. zeylanicum* essential oil significantly decreased the concentration of MDA in plasma and duodenal mucosa, however it had no significant effect on the concentration of MDA in the liver and kidney tissues. The dose of cinnamon oil induced the SOD production in liver and heart tissues. Moreover, there was significant interaction between enviromental condition and dose of cinnamon oil in the liver tissue. GSH levels of heart and kidney and GSH-Px activity of heart were also found higher in cinnamon supplemented groups under different enviromental condition. These findings indicated that cinnamon oil had protective effects on liver, heart and kidney by activating antioxidant mechanism in the cells. In agree with these results, MOSELHY and JUNBI [38] reported that cinnamon contained high level of phenolic groups had a potent hepatoprotective activity by inhibiting the chain reaction of lipid peroxidation resulting decrease in MDA level and elevate in SOD activities. ULLAH et al.[39] showed that *C. zeylanicum* had strong nephroprotective effect, especially against aminoglycosides induced nephrotoxicity due to its strong antioxidant property. NOORI et al.[40] reported that

cinnamon markedly showed antioxidant activity in liver, kidney and heart tissues. In addition, cinnamon is rich from lots of vitamins and minerals [11,16]. These contents could be effective on antioxidant mechanism of cinnamon and cause well-being of the birds.

The results obtained from this study have given some clues on the chemotaxonomy of this plants. *Cinnamaldehyde* is predominant compound in essential oil of *Cinnamomum zeylanicum* bark. Chronic heat stress increased oxidative stress in hepatic, heart and renal tissues, which is characterized by reduction of the antioxidant enzyme activities and glutathione levels, and it resulted with the elevation of MDA level. Both doses of *C. zeylanicum* bark oil exhibited significant antioxidant activity in growing quails through the activation of antioxidant enzymes, especially under heat stress condition.

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