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Research Article

# The Effects of Dietary Lead Exposure and Ascorbic Acid on Performance, Lipid Peroxidation Status and Biochemical Parameters of Broilers*

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**Abstract:** Lead is one of the ubiquitous environmental pollutants that induce a broad range of physiological and biochemical dysfunctions in animals. This study evaluated the effects of dietary lead exposure and ascorbic acid on performance, serum biochemical parameters, plasma malondialdehyde and lead accumulation in broiler chickens. For this purpose, lead acetate at 200 mg/kg and ascorbic acid at 100 mg/kg were added to the diet alone or in combination for 42 days. A total of 120 broiler chicks were divided into 4 treatment groups: control, ascorbic acid, lead, and lead + ascorbic acid. By the end of the study, lead caused body weight and body weight gain to decrease significantly, although its effects on feed consumption and the feed conversion ratio were not significant. While lead did not alter the serum lactate dehydrogenase, aspartate aminotransferase or alanine aminotransferase activities or albumin or total protein concentrations, it increased malondialdehyde ($P < 0.001$) and triglyceride ($P < 0.01$) levels. Although the lead contents of the serum and muscle were unchanged, lead was accumulated in the liver and kidneys ($P < 0.001$). Our results showed that lead (200 mg/kg diet) had an inhibitory effect on the growth of broilers and appeared to be inducing lipid peroxidation. The addition of ascorbic acid to the diet reduced the plasma malondialdehyde levels induced by lead and tended to reduce the inhibitory effect of lead on growth. It is concluded that the addition of higher doses of ascorbic acid to the diet may be more efficacious in fully reversing the negative effect of lead on growth.

**Key Words:** Broilers, lead, ascorbic acid, performance, biochemical parameters, lipid peroxidation

## Kurşun ve Askorbik Asidin Broyler Piliçlerde Besi Performansı, Lipit Peroksidasyon Düzeyi ve Biyokimyasal Parametreler Üzerine Etkisi

**Özet:** Kurşun hayvanlarda fizyolojik ve biyokimyasal fonksiyon bozukluluklarına yol açan, yaygın bulunan çevre kirleticilerden biridir. Bu araştırmada broyler piliçlerin yemle kurşuna maruz kalmasının ve askorbik asit ilavesinin performans, serum biyokimyasal parametreler ile plazma malondialdehid seviyesi ve kurşun birikim düzeyi üzerine etkisi belirlendi. Bu amaçla, kurşun asetat 200 mg/kg ve askorbik asit 100 mg/kg oranında ayrı ayrı ve birlikte 42 gün süreyle karma yeme katıldı. Araştırmada 120 adet broyler civciv 3'er alt gruplu: kontrol, askorbik asit, kurşun ve kurşun + askorbik asitten oluşan 4 gruba ayrıldı. Araştırma sonunda kurşun ilavesi, canlı ağırlık ve canlık ağırlık artışında belirgin düşüklüğe neden oldu ($P < 0,05$). Yem tüketimi ve yemden yararlanma oranı, kurşun ve askorbik asidin ayrı ayrı ve birlikte katılmasından etkilenmedi. Kurşun, serum laktat dehidrojenaz, aspartat aminotransferaz, alanin aminotransferaz aktivitesi, albumin ve total protein konsantrasyonunu etkilemezken, malondialdehid ($P < 0.001$) ve trigliserid ($P < 0,01$) seviyesini belirgin olarak yükseltti. Serum ve kas kurşun düzeyi değişmezken, kurşun, böbrek ve karaciğerde birikim yaptı ($P < 0,01$). Bu araştırma sonucunda, kurşunun (200 mg/kg yem) broylerlerde büyümeyi baskıladığı ve plasma malondialdehid seviyesini yükselttiği belirlenmiştir. Yeme katılan askorbik asit kurşunun neden olduğu lipid peroksidasyonunu düşürmüş ve kurşunun büyümeyi baskılayıcı etkisini azaltma eğiliminde olduğu belirlenmiştir. Rasyona daha yüksek dozlarda askorbik asit katılmasının kurşunun büyüme üzerine olan olumsuz etkisinin tamamen giderilmesinde etkili olabileceği sonucuna varılmıştır.

**Anahtar Sözcükler:** Broyler, kurşun, askorbik asit, performans, biyokimyasal parametreler, lipit peroksidasyonu

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## Introduction

Lead is one of the ubiquitous environmental pollutants, particularly widespread in industrial areas. Animals are exposed to lead from numerous sources as well as from the general environment. The main sources of contamination of feed by lead are soil, industrial pollution, agricultural technology and feed processing. Ingested lead has resulted in poisoning, poor performance and death in animals (1,2).

Such accumulated lead is toxic in most of its chemical forms, whether it is inhaled or ingested in water or feed. The extent to which orally administered lead is absorbed is small. However, due to its slow rate of elimination, harmful levels of lead can accumulate in tissues after prolonged exposure to low quantities (3,4). Lead produces acute and chronic poisoning and induces a broad range of physiological, biochemical and behavioral dysfunctions in animals. Indeed, many enzymes, membranes and biochemical processes have been shown to be affected by lead, but none has been shown to be both sensitive and of key importance in explaining the manifestation of toxicity. Among the well-established effects of lead is anemia due to inhibition of heme synthesizing enzymes. Lead is also known to reduce erythrocyte membrane stability, while the formation of intranuclear inclusion bodies is one of the earliest manifestations of exposure to lead. Lead affects the metabolism of other minerals and has an affinity for bone, where it acts by replacing calcium; thus the highest concentrations of lead are usually found in bone, kidney and liver (5-7).

Studies have reported that lead has a potential for inducing oxidative stress and acts as a catalyst in the oxidative reactions of biological macromolecules. Therefore, the toxicities associated with this metal might be due to oxidative tissue damage (2,4,8-10). To prevent peroxidative tissue damage, there are protective mechanisms in vivo, such as an enzymatic defense system (antioxidant enzymes) and free-radical scavengers (antioxidants). Ascorbic acid is a well-known antioxidant vitamin involved in several biochemical processes in biological systems. This vitamin breaks the chain of lipid peroxidation in cell membranes and scavenges free radicals such as reactive oxygen species (11,12).

Although lead toxicity has been extensively studied in animals, there are only a limited number of studies on the effects of lead on broiler performance. Therefore, the purpose of the present study was to evaluate the effects of dietary lead exposure on performance, biochemical parameters, lipid peroxidation status and lead concentrations in the serum, muscle, liver and kidneys of broilers. We also aimed to determine whether the negative effects of lead, attributed to tissue peroxidation, could be reversed by adding ascorbic acid.

## Materials and Methods

### Animal, feed and experimental design

One hundred twenty 1 day-old Ross broiler chicks of both sexes were used. Individually weighed chicks were randomly assigned to 4 treatment groups in 3 replicates of 10 birds each. The chicks were fed a basal starter diet until 21 days of age. This was followed by a basal grower-finisher diet from day 21 to day 42. The ingredients and the chemical composition of the diets are shown in Table 1. The diets were formulated according to National

Table1. Ingredients and chemical composition of the basal diets.

| | Starter Diet, % | Grower Diet, % |
|---|---|---|
| Corn | 45.00 | 43.00 |
| Wheat | 5.00 | 11.00 |
| Wheat bran | 2.00 | 4.00 |
| Soybean meal | 33.00 | 27.35 |
| Fish meal | 6.00 | 4.00 |
| Vegetable oil | 6.00 | 7.00 |
| Limestone | 1.30 | 1.50 |
| Dicalcium phosphate | 0.65 | 1.00 |
| Salt | 0.30 | 0.30 |
| Vitamin premix[1] | 0.30 | 0.30 |
| Mineral premix[2] | 0.30 | 0.30 |
| DL-Methionin | 0.15 | 0.15 |
| Chemical analysis | | |
| Dry matter | 89.50 | 89.00 |
| Crude protein | 23.00 | 20.00 |
| Ether extract | 8.50 | 9.00 |
| Ash | 6.50 | 7.00 |
| Calculated values | | |
| Metabolizable energy (MJ/kg) | 12.97 | 13.38 |
| Calcium | 0.91 | 1.00 |
| Phosphorus | 0.65 | 0.65 |

1: 15,000,000 IU vitamin A, 3,000,000 IU cholecalciferol, 70 g vitamin E, 5 g menadion, 3 g thiamin, 6 g riboflavin, 5 g pyridoxin, 0.03 g vitamin $B_{12}$, 0.075 g biotin, 50 g ascorbic acid , 25 g niacin, 200 g choline chloride, 1 g folic acid, 12 g Ca-D-pantothenate/2 kg premix.
2: 80 g Mn, 60 g Fe, 60 g Zn, 5 g Cu, 0.20 g Co, 1 g I, 0.150 g Se/1 kg premix

Research Council (13) guidelines. Chemical compositions of the diets were analyzed as recommended by the Association of Official Analytical Chemists (AOAC) (14).

Lead was added to the diet at a level of 200 mg/kg, since the toxic dietary concentration of lead was reported at between 1000 and 200 mg/kg for chickens (13). The dietary supplemental level of ascorbic acid has varied in different studies (11,13). In the present study, ascorbic acid was added to the diet at a level of 100 mg/kg. The experimental design consisted of 4 dietary treatments: (1) control: basal diet; (2): ascorbic acid 100 mg/kg basal diet; (3): lead (as lead acetate $x3H_20$) 200 mg/kg basal diet; (4): ascorbic acid 100 mg plus lead 200 mg/kg basal diet.

Birds were weighed at weekly intervals during the experimental period to determine body weight (BW) and body weight gain (BWG). Feed consumption (FC) was measured weekly. Feed conversion ratio (FCR) was calculated as feed-to-gain ratio, on the $7^{th}$, $14^{th}$, $21^{st}$, $28^{th}$, $35^{th}$ and $42^{nd}$ days and over days 1 to 42 of the experiment. The chicks were housed in electrically heated batteries under fluorescent lighting and allowed ad libitum access to feed and water. The animal care and use protocol was reviewed and approved by the Ethical Committee of the Faculty of Veterinary Medicine, Mustafa Kemal University.

### Serum biochemical analyses

When the chicks were 6 weeks of age, the feeding trial finished and blood samples were collected from 12 broilers randomly chosen (4 chicks per replicate) from each group. Serums were separated and immediately analyzed for lactate dehydrogenase (LDH), aspartate aminotransferase (AST), alanine aminotransferase (ALT) activities and total protein and albumin levels as an indicator of liver functions. In addition, serum triglyceride concentrations were measured on an analyzer (AMS, Rome, Italy) using diagnostic kits (Teco Diagnostics, CA, USA). Plasma MDA levels were determined for lipid peroxidation status using the spectrophotometric method described by Yoshioko et al. (15).

### Serum, muscle, liver and kidney lead contents

Tissue samples were collected immediately after slaughtering and transported to the laboratory. Visible fat and connective tissue on the tissue samples were removed and frozen separately in plastic bags and stored at –20 °C until analyzed. Serum samples were also stored at –20 °C until analyzed. Tissue and serum samples were treated with nitric acid and hydrochloric acid digestion according to the methods of Alonso et al. (16) and Lai and Jamieson (17), respectively, for lead analysis at 283.306 nm by inductively coupled plasma-atomic emission spectrometry (ICP-AES, Liberty Series-II Varian, USA).

### Statistical Analysis

The data between groups for performance parameters and all biochemical values were subjected to one-way analysis of variance (ANOVA) using a statistical analyses system configured for the computer (18). Where appropriate, post-hoc analyses were carried out using Duncan's test. All statements of significance are based on the 0.05 level of probability.

## Results

The effects of lead and ascorbic acid on BW and BWG and FC and FCR are presented in Tables 2 and 3, respectively. Supplemental dietary lead significantly reduced BW and BWG ($P < 0.05$) but its effects on FC ($P > 0.05$) and FCR were not significant. No clinical signs of lead toxicity were observed in the broilers administered lead. Mortality rates of the groups are given in Table 2. The deaths in the groups were trauma related.

Serum biochemical values are shown in Table 4. Lead supplementation increased the plasma MDA ($P < 0.001$) and serum triglyceride ($P < 0.01$) levels. No significant changes were observed in serum albumin, total protein levels or LDH, AST or ALT activities ($P > 0.05$). Lead concentrations in serum, liver, kidneys and muscle are given in Table 5. Lead was accumulated in the liver ($P < 0.05$) and kidney ($P < 0.01$) but not in the serum or muscle.

## Discussion

Lead addition to the diet at 200 mg/kg reduced growth in terms of BW (Table 2) and BWG (Table 3) at the end of the study ($P < 0.05$). BWG was statistically lower in the lead-treated group than that in the control and ascorbic acid-treated groups ($P < 0.05$) while the BWG of the lead-plus-ascorbic acid group was similar to that in the control and lead-treated groups at the end of the $6^{th}$ week. Thus ascorbic acid addition to diet tended to reverse the depressive effect of lead. However, the negative effect of lead was not fully overcome by the

Table 2. The effect of lead (200 mg/kg) and ascorbic acid (100 mg/kg) on body weight (g) of broilers.

| | Treatment Groups | | | | | | | | |
|---|---|---|---|---|---|---|---|---|---|
| | Control | | Ascorbic acid | | Lead | | Lead + Ascorbic acid | | |
| Day | n | x ± Sx | n | x ± Sx | n | x ± Sx | n | x ± Sx | F |
| 1 | 30 | 41.57 ± 0.72 | 30 | 41.43 ± 0.92 | 30 | 41.23 ± 0.70 | 30 | 41.10 ± 0.71 | 0.07 |
| 7 | 30 | 153.17 ± 3.71 | 30 | 151.77 ± 2.85 | 30 | 145.40 ± 2.76 | 30 | 146.37 ± 3.35 | 1.47 |
| 14 | 30 | 366.83 ± 9.27 | 30 | 362.10 ± 6.84 | 30 | 351.80 ± 8.86 | 30 | 347.27 ± 11.43 | 0.95 |
| 21 | 30 | 737.43 ± 13.43 | 30 | 734.30 ± 14.97 | 29 | 696.42 ± 13.35 | 30 | 719.70 ± 14.22 | 1.76 |
| 28 | 30 | 1239.20 ± 25.09 | 29 | 1232.48 ± 18.52 | 29 | 1176.14 ± 19.33 | 30 | 1221.23 ± 22.90 | 1.69 |
| 35 | 29 | $1709.00^{a}$ ± 26.69 | 29 | $1726.03^{a}$ ± 33.48 | 28 | $1622.46^{b}$ ± 28.88 | 30 | $1655.27^{b}$ ± 26.21 | 2.70* |
| 42 | 29 | $2187.17^{a}$ ± 34.72 | 29 | $2238.28^{a}$ ± 45.88 | 28 | $2065.32^{b}$ ± 38.44 | 29 | $2127.93^{b}$ ± 36.38 | 3.62* |

$^{a,b}$: Means within a row with no common superscript differ significantly (*P < 0.05).

Table 3. The effects of lead (200 mg/kg) and ascorbic acid (100 mg/kg) on feed consumption (g), body weight gain (g) and feed conversion ratio (g of feed/g of live weight gain) of broilers.

| Day | | Treatment Groups | | | | |
|---|---|---|---|---|---|---|
| | | Control | Ascorbic Acid | Lead | Lead + Ascorbic Acid | |
| | | x ± Sx | x ± Sx | x ± Sx | x ± Sx | F |
| 01-7 | Feed consumption | 156.00 ± 7.29 | 149.35 ± 4.73 | 149.49 ± 4.26 | 148.63 ± 7.30 | 0.32 |
| | Body weight gain | 111.60 ± 3.61 | 110.33 ± 3.97 | 104.07 ± 3.12 | 105.30 ± 1.40 | 1.35 |
| | Feed conversion ratio | 1.4 | 1.36 | 1.41 | 1.41 | |
| 8-14 | Feed consumption | 288.3 ± 3.83 | 292.37 ± 2.99 | 289.10 ± 10.32 | 293.30 ± 13.03 | 0.08 |
| | Body weight gain | 213.67 ± 10.68 | 210.34 ± 2.58 | 206.40 ± 3.09 | 200.9 ± 7.25 | 0.42 |
| | Feed conversion ratio | 1.36 | 1.39 | 1.45 | 1.45 | |
| 15-21 | Feed consumption | 519.68 ± 15.77 | 532.04 ± 22.84 | 522.79 ± 15.64 | 537.42 ± 9.74 | 0.24 |
| | Body weight gain | 370.60 ± 28.74 | 372.06 ± 12.65 | 343.45 ± 16.64 | 372.43 ± 8.03 | 0.63 |
| | Feed conversion ratio | 1.42 | 1.43 | 1.44 | 1.44 | |
| 22-28 | Feed consumption | 884.33 ± 10.73 | 819.36 ± 30.60 | 823.33 ± 14.53 | 833.92 ± 21.96 | 2.07 |
| | Body weight gain | 501.77 ± 17.43 | 497.56 ± 5.79 | 479.83 ± 2.11 | 502.31 ± 20.58 | 0.58 |
| | Feed conversion ratio | 1.77 | 1.65 | 1.67 | 1.67 | |
| 29-35 | Feed consumption | 970.35 ± 19.87 | 1011.41 ± 33.22 | 984.46 ± 28.11 | 911.29 ± 20.55 | 2.64 |
| | Body weight gain | 469.70 ± 19.84 | 493.37 ± 9.51 | 446.81 ± 27.40 | 433.26 ± 22.92 | 1.59 |
| | Feed conversion ratio | 2.08 | 2.05 | 2.11 | 2.11 | |
| 36-42 | Feed consumption | 966.33 ± 36.44 | 999.15 ± 30.62 | 996.93 ± 68.96 | 1002.33 ± 19.38 | 0.15 |
| | Body weight gain | 479.50 ± 19.50 | 511.51 ± 28.42 | 442.42 ± 30.15 | 466.21 ± 13.30 | 1.46 |
| | Feed conversion ratio | 2.02 | 1.96 | 2.15 | 2.15 | |
| 1-42 | Feed consumption | 3785.01 ± 79.02 | 3803.69 ± 78.19 | 3766.10 ± 84.73 | 3726.90 ± 46.65 | 0.2 |
| | Body weight gain | $2146.83^{ab}$ ± 21.59 | $2195.31^{a}$ ± 39.63 | $2023.08^{c}$ ± 36.90 | $2080.41^{bc}$ ± 8.36 | 6.56* |
| | Feed conversion ratio | 1.76 | 1.73 | 1.79 | 1.79 | |

$^{a-c}$: Means within a row with no common superscript differ significantly (*P < 0.05).

Table 4. The effect of lead (200 mg/kg) and ascorbic acid (100 mg/kg) on selected biochemical parameters in serum and MDA levels in plasma.

| | Treatment Groups | | | | |
|---|---|---|---|---|---|
| | Control | Ascorbic acid | Lead | Lead + Ascorbic acid | |
| | x ± Sx | x ± Sx | x ± Sx | x ± Sx | F |
| MDA, (IU/l) | 4.95[a] ± 0.75 | 5.79[a] ± 0.03 | 26.05[c] ± 2.09 | 8.81[b] ± 0.32 | 101.02*** |
| Albumin, (g/dl) | 0.97 ± 0.03 | 1.0 ± 0.03 | 1.0 ± 0.04 | 1.0 ± 0.04 | 0.6 |
| Total protein, (g/dl) | 2.3 ± 0.05 | 2.4 ± 0.05 | 2.5 ± 0.05 | 2.4 ± 0.09 | 1.67 |
| Triglyceride, (mg/dl) | 35.8[a] ± 4.8 | 31.8[a] ± 1.7 | 64.33[b] ± 6.0 | 39.5[a] ± 7.3 | 6.73** |
| LDH, (IU/l) | 550.8 ± 44.3 | 500.7 ± 39.3 | 520.0 ± 44.1 | 438.8 ± 29.5 | 1.57 |
| AST, (IU/l) | 210.2 ± 22.6 | 221.0 ± 25.2 | 203.6 ± 13.9 | 190.8 ± 11.1 | 0.37 |
| ALT, (IU/l) | 25.2 ± 3.2 | 29.7 ± 2.9 | 29.4 ± 4.9 | 29.7 ± 1.6 | 0.44 |

[a-c]: Means within a row with no common superscript differ significantly (**P < 0.01,***P < 0.001).

Table 5. The lead contents of serum and selected tissues.

| | Treatment Groups | | | | |
|---|---|---|---|---|---|
| | Control | Ascorbic acid | Lead | Lead + Ascorbic acid | |
| | x ± Sx | x ± Sx | x ± Sx | x ± Sx | F |
| Serum (mg/l) | 0.03 ± 0.00 | 0.03 ± 0.00 | 0.04 ± 0.00 | 0.02 ± 0.00 | 1.63 |
| Liver (mg/kg) | 0.92[a] ± 0.23 | 0.98[a] ± 0.24 | 1.71[b] ± 0.14 | 1.47[ab] ± 0.19 | 3.52* |
| Kidney (mg/kg) | 1.31[a] ± 0.21 | 1.19[a] ± 0.18 | 2.33[b] ± 0.25 | 2.33[b] ± 0.40 | 5.31** |
| Muscle (mg/kg) | 0.71 ± 0.14 | 0.63 ± 0.18 | 0.41 ± 0.15 | 0.60 ± 0.20 | 0.54 |

[a-b]: Means within a row with no common superscript differ significantly
(*P < 0.05, **P < 0.01), according to Duncan's multiple range tests.

addition of ascorbic acid at a dose of 100 mg/kg of diet. It may be said that 100 mg ascorbic acid/kg of diet was insufficient for full recovery and a higher dose of ascorbic acid in the diet would be more effective in overcoming the growth inhibitory effect of lead. Although the effects of lead and ascorbic acid on FC (Table 3) and FCR (Table 3) were not significant, in the lead group, FC was 5.7% lower than that in the control group, while the FCR was highest in this group. Previous studies have reported decreased weight gain and feed intake and an increased feed:gain ratio in lead exposed chickens (19,20). Bakalli et al. (20) found that even 1 mg of lead/kg of feed significantly depressed body gains and that significant negative effect of lead on FCR were found at a level of 10 mg of lead/kg of feed. The findings of that study in terms of FCR are not in agreement with those of the present study. Lead at 200 mg/kg of diet was found to have no significant effect on FCR in the present study. It is known that there are many different factors affecting the quantity of lead needed to produce toxicity. This difference might be associated with factors such as the nature of the compound in which it is present, diet composition or the presence of other minerals in the diet, duration and animal susceptibility, which others have shown to affect the degree of lead toxicity in animals (5,13,21). Similar to the results of the present study, Vodela et al. (22) reported that the addition of a heavy metal mixture (including lead) to broiler drinking water

caused growth to decrease. Growth depression may be due to metabolic disorders associated with lead, such as inhibition of enzymes involved in heme synthesis and the oxidase system. Lead also has a strong affinity to mitochondria and many of its pathological effects may be caused by ultra structural and functional changes in these organelles (2,6,21). It has been demonstrated that lead has a potential for inducing oxidative stress, which may in turn result in loss of cellular functions and tissue damage, possibly leading to growth depression and impairment of health (8,9,23).

Lead significantly altered plasma MDA and triglyceride concentrations (Table 4). MDA is known as an indicator for lipid peroxidation associated with the oxidation of polyunsaturated fatty acids (2,9). This result is in agreement with that of Yiin and Lin (9), who showed that in vitro incubation of polyunsaturated fatty acids with lead at 37 °C for 24 h markedly increased MDA levels. The results of that in vitro study indicated that the important mechanism of the acute toxic effects of lead compounds is due, at least in part, to metal-catalyzed peroxidation of polyunsaturated fatty acids. In the present study, the elevated MDA level was reduced by ascorbic acid addition to the diet containing lead.

The highest serum triglyceride level was observed in the lead treated group ($P < 0.01$). This increase in triglyceride levels might have been due to enhanced metabolic stress induced by lead. Ascorbic acid addition significantly reduced the lead-enhanced triglyceride level. It has been postulated that ascorbic acid alleviates the effect of metabolic stress (12). Lead had no significant effects on serum albumin, total protein levels or LDH, AST or ALT activities which findings were similar to those of Khan et al. (21). Khan et al. (24) also reported that subacute lead toxicity increased AST activity, but did not affect total protein levels or ALT activity.

Supplementation with dietary lead did not change serum or muscle lead concentrations (Table 5). However, lead was significantly accumulated in the liver and kidneys of lead-treated broilers compared to the control and ascorbic acid-treated groups. This is in agreement with previous reports that lead concentrations in the liver and kidneys may be high in animals that have no toxication symptoms and a normal blood lead level (3,5). Further support for our findings comes from Khan et al. (25, 26), who report that toxic doses of lead administrated orally accumulated in the liver.

The current study showed that lead has a growth depressive effect although FCR was not affected at 200 mg of lead/kg of diet during the commercial rearing period of 42 days. Lead increased plasma MDA concentrations which are evidence of lipid peroxidation. Ascorbic acid supplementation reduced plasma MDA and triglyceride levels induced by lead and the negative effect of lead on growth. It was concluded that further studies should be carried out to show the fully reversing effect on growth depression caused by lead of ascorbic acid at higher doses. In addition, lead is entirely accumulated in the kidneys and liver but not in broiler meat per se.

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## References

1. McDowell, L.S.: Minerals in Animal and Human Nutrition. Academic Press, Inc, California. 1992; 361-364.
2. Gurer, H., Ercal, N.: Can antioxidants be beneficial in the treatment of lead poisoning. Free Radical Bio. Med., 2000; 29: 927-945.
3. Demichele, S.J.: Nutrition of lead. Comp. Biochem. Phys. Part A: Physiology, 1984; 78, 401-408.
4. Ercal, N., Gurer-Orhan, H., Aykin-Burns, N.: Toxic metals and oxidative stress. Part I: Mechanisms involved in metal-induced oxidative damage. Curr. Top. Med. Chem., 2001; 1: 529-539.
5. Humphreys, D.J.: Effects of exposure to excessive quantities of lead on animals. Br. Vet. J., 1991; 147: 18-30.
6. Mertz, W.: Trace Elements in Human and Animal Nutrition. 5th Ed. Academic Press Inc., Florida, 1986; 281-307.
7. Heaney, R.P.: Lead in calcium supplements. J. Am. Med. Assoc., 2000; 284: 1432-1433.
8. Donaldson, W.E.: Interactions of dietary lead with fish oil and antioxidant in chicks. Biol. Trace Elem. Res., 1991; 31: 215-222.

9. Yiin, S., Lin, T.H.: Lead-catalyzed peroxidation of essential unsaturated fatty acid. Biol. Trace Elem. Res., 1995; 50: 167-172.

10. Galaris, D., Evangelou, A.: The role of oxidative stress in mechanisms of metal-induced carcinogenesis. Crit. Rev. Oncol. / Hemat., 2001; 42: 93-103.

11. Carr, A., Frei, B. : Does vitamin C act as a pro-oxidant under physiological conditions? FASEB J., 1999; 13: 1007-1024.

12. Kucuk, O., Sahin, N., Sahin, K., Gursu, M.F., Gulcu, F., Ozcelik, M., Issi, M.: Egg production, egg quality, and lipid peroxidation status in laying hens maintained at a low ambient temperature (6 ºC) and fed a vitamin C and vitamin E-supplemented diet. Vet. Med.-Czech., 2003; 1-2: 33-40.

13. National Research Council (NRC): Nutrient Requirements of Poultry: 9th Rev. Ed. National Academy Press, Washington, DC. 1994; 44-45.

14. A.O.A.C.: Official Methods of Analysis. 15th Ed. Association of Official Analytical Chemists. Arlington, VA, USA. 1990.

15. Yoshioko, T., Kawada, K., Shimada, T.: Lipid peroxidation in maternal and cord blood and protective mechanism against activated-oxygen toxicity in the blood. Am. J. Obstet. Gynecol., 1979; 135: 372-376.

16. Alonso, M.L., Benedito, J.L., Miranda, M., Castillo, C., Hernandez, J., Shore, R.F.: Arsenic, cadmium, lead, copper and zinc in cattle from Galicia, NW Spain. Sci. Total Environ., 2000; 246: 237-248.

17. Lai, J.J., Jamieson, G.C.: Determination of dysprosium in monkey serum by inductively coupled plasma atomic emission spectrometry after the administration of Sprodiamide injection, a new contrast medium for magnetic resonance imaging. J. Pharmaceut. Biomed., 1993; 11: 1129-1134.

18. SPSS Inc.: SPSS for Windows. Base System User's Guide, Release 9.05. SPSS Inc., Chicago. 1998.

19. Cupo, M.A., Donaldson, W.E.: Effect of lead and niacin on growth and serotonin metabolism in chicks. J. Nutr., 1988; 118: 107-113.

20. Bakalli, R.I., Pesti, G.M., Ragland, W.L.: The magnitude of lead toxicity in broiler chickens. Vet. Hum. Toxicol., 1995; 37: 15-19.

21. Khan, M.Z., Szarek, J., Koncicki, A., Krasnodebska-Depta, A.: Oral administration of monensin and lead to broiler chicks: effects on haematological and biochemical parameters. Acta Vet. Hung., 1994; 42: 111-120.

22. Vodela, J.K., Lenz, S.D., Renden, J.A., McElhenney, W.H., Kemppainen, B.W.: Drinking water contaminants (arsenic, cadmium, lead, benzene and trichloroethylene). 1. Interaction of contaminants with nutritional status on general performance and immune function in broiler chickens. Poult. Sci., 1997; 76: 1474-1492.

23. Donaldson, W.E., Leeming, T.K.: Dietary lead: effects on hepatic fatty acid composition in chicks. Toxicol. Appl. Pharmacol., 1984; 73: 119-123.

24. Khan, M.Z., Szarek, J., Krasnodebska-Depta, A., Koncicki, A.: Effects of concurrent administration of lead and selenium on some hematological and biochemical parameters of broiler chickens. Acta Vet. Hung., 1993; 41: 123-137.

25. Khan, M.Z., Szarek, J., Markiewicz, K.: Effects of oral administration of toxic levels of lead and selenium upon concentration of different elements in the liver of broiler chicks. Zentralbl. Veterinarmed. A, 1993; 40: 652-664.

26. Khan, M.Z., Szarek, J., Markiewicz, K., Markiewicz, E.: Effects of concurrent oral administration of toxic levels of monensin and lead on concentration of different elements in the liver of broiler chicks. Zentralbl. Veterinarmed. A, 1993; 40: 466-475.