*Iraqi Journal of Veterinary Sciences, Vol. 25, No. 1, 2011 (1-7)*

# Changes in some blood parameters in lactating female rats and their pups exposed to lead: effects of vitamins C and E

**H. M. Jassim and A. A. Hassan**

Department of Physiology, Biochemistry and Pharmacology, College of Veterinary Medicine, University of Mosul, Mosul, Iraq

(Received February 17, 2009; Accepted April 15, 2010)

**Abstract**

The present work was designed to examine the changes in some blood parameters in lactating rats treated with lead acetate (10 mg /kg B.W. orally) and its interaction with vitamin E (600 mg/kg diet) or vitamin C (100 mg/kg B.W. orally) during lactation period (20 days) and their pups. Administration of lead acetate to the female lactating rats caused a significant decrease in packed cell volume (PCV), hemoglobin concentration (Hb), red blood cell count (RBC), body weight, mean corpuscular heamoglobin concentration (MCHC) whereas the white blood cells count (WBC), total proteins, the percentage of monocyte and mean corpuscular volume(MCV) significantly increased administration of lead acetate to female lactating rats produced a significant decrease in PCV, Hb, RBC, MCHC, body weight, and the percentage of the neotrophils in their pups. But the WBC count, total proteins, the percentage of lymphocyte, monocyte, MCV a significantly increased in their pups. Treatment dams with vitamin E concomitantly with lead acetate increased the PCV, Hb, MCHC, whereas percentage of monocyte significantly decreased, PCV, Hb, RBC, the percentage of neutrophils a significantly increased, whereas WBC count, the percentage of lymphocyte decreased significantly in their pups of this group of dams. Treatment dams with vitamin C concomitantly with lead acetate significantly increased the PCV, MCV, whereas percentage of monocytes significantly decreased, but Hb, PCV and RBC significantly increased in their pups. It could be concluded that treatment female lactating rats with vitamin E or C concomitantly with lead acetate exert an antioxidant effect on blood constituent in dams and their pups and vitamin E more effective than vitamin C.

***Keywords:*** Lead acetate; Lactating rats; Pups; Complete blood picture.
Available online at http://www.vetmedmosul.org/ijvs

# التغيرات في بعض مكونات الدم في إناث الجرذان المرضعات وصغارها المعرضة للرصاص: تاثيري فيتامين هـ وجـ

**هبة محمد جاسم و أشواق احمد حسن**

فرع الفسلجة والكيمياء الحياتية والادوية، كلية الطب البيطري، جامعة الموصل، الموصل، العراق

**الخلاصة**

صممت تجارب البحث الحالي لدراسة التغيرات في بعض معايير الدم في الجرذان المرضعات المعاملة بخلات الرصاص (١٠ملغم/كغم من وزن الجسم عن طريق الفم) وتداخله مع فيتامين هـ (600 ملغم /كغم عليقه) او فيتامين جـ (100 ملغم /كغم وزن الجسم عن طريق الفم) خلال فترة الرضاعة (٢٠ يوم) وصغارها. سبب إعطاء خلات الرصاص لإناث الجرذان المرضعات انخفاض معنوي في حجم الخلايا المرصوصة وتركيز الهيموكلوبين والعدد الكلي لخلايا الدم الحمراء ووزن الجسم ومعدل تركيز هيموكلوبين الكرية بينما احدثت المعاملة زيادة معنوية في العدد الكلي لخلايا الدم البيضاء والبروتينات الكلية والنسب المئوية للخلايا وحيدة النواة ومعدل حجم الكريه. احدث إعطاء خلات الرصاص إلى إناث الجرذان المرضعات انخفاض معنوي في حجم الخلايا المرصوصة وتركيز الهيموكلوبين والعدد الكلي لخلايا الدم الحمراء ومعدل تركيز هيموكلوبين الكريه وأوزان الجسم والنسبة المئوية للعدلات في

صغارها، لكن عدد خلايا الدم البيضاء والبروتينات الكلية ومعدل حجم الكريه والنسب المئوية للخلايا اللمفية والخلايا وحيدة النواة كانت مرتفعة معنويا في صغارها. وسببت معاملة الامهات بفيتامين هـ سوية مع خلات الرصاص زيادة معنوية في حجم خلايا الدم المرصوصة وتركيز الهيموكلوبين ومعدل تركيز هيموكلوبين الكريه، بينما نسب الخلايا وحيدة النواة كانت منخفضه معنويا. حجم خلايا الدم المرصوصة وتركيز الهيموكلوبين والعدد الكلي لخلايا الدم الحمراء والنسب المئوية للعدلات ازدادت معنويا,بينما العدد الكلي لخلايا الدم البيضاء والنسب المؤية للخلايا اللمفية انخفضت معنويا في صغار هذه المجموعة. أحدثت معاملة الامهات بفيتامين جـ سوية مع خلات الرصاص زيادة معنوية في حجم خلايا الدم المرصوصة ومعدل حجم الكريه بينما النسب المئوية للخلايا وحيدة النواة انخفضت معنويا، لكن تركيز الهيموكلوبين وحجم خاليا الدم المرصوصة والعدد الكلي لخلايا الدم الحمراء ارتفعت معنويا في صغارها. وقد استنتج من الدراسة أن معاملة إناث الجرذان المرضعات بفيتامين هـ أو جـ سوية مع خلات الرصاص قد احدث تأثيرا مضادا للاكسدة على مكونات الدم في الأمهات وصغارها, وان تأثير فيتأمين هـ أكثر فعالية من فيتأمين جـ.

## Introduction

Lead is non-essential heavy metal widely distributed in the environment, chronic exposure to low levels of this agent is one of the problems of public health, due to its toxicity, Source of lead exposure may include air, water, food, soil (1), breast milk can however be pathway of maternal excretion of lead and this toxin impact most severely on the newborn (2,3). Lead produce toxicological and pathological effects on central nervous system, peripheral nerves, kidneys and hematopoitic system (1), as well as infertility (4). Lead is known to induce oxidative stress leading to tissue damage (5), and lipid peroxidation by inhibiting the synthesis of some antioxidant enzymes such as phospholipids hydroperoxidase glutathione peroxidase (PHGPX) (6).

It has long been known that heamatopoiesis and heme synthesis affected by lead poisoning (1).

Lead can cause damage in the erythrocyte, originating defective cells, preventing them from carrying oxygen also it produce high blood pressure that increases the risk of heart attack (7).

An increase in oxidative damage to body cells and tissues is balanced by powerful enzymatic and non enzymatic antioxidant defense system that scavenge and suppress the formation of reactive oxygen species (ROS) and protect cells from lipid peroxide – mediated damage (8) e.g. (Carotinoids, vitamin E & C, Flavonoids, GSH-Peroxidase, Soperoxide – dismutase, Catalase) (9). Vitamins C&E are great powerful antioxidants, protect cells from oxidation, vitamin C is biological reducing agent, vitamin E act by giving up of its electrons to the electron deficient free radicals making it more stable (10). Both vitamins C & E helping to prevent degenerative disease such as cardiovascular diseases (10).

## Materials and methods

Twenty female albino rats (dams) obtained from the animal house of Veterinary Medical College, University of Mosul, at (3-4) months of age, they were housed in polypropylene cages under controlled conditions of temperature (18-20) c° and lighting (10 hours light /14 hours dark). The dams were supplied a standard pellet diet and tap water *ad libitum*.

The dams and their pups were randomly divided at the first day of parturition in to four groups (5 dams /group). The first group received tap water (1ml/kg BW) during lactation period(20 days) serve as control. The second group received lead acetate (10 mg /kg B.W. orally) during lactation period (20 days), lead acetate dissolved in distalled water and given at volume of 1ml/kg (4). The third group received lead acetate (10 mg /kg B.W) concomitantly with feeding of vitamin E (600 mg / kg diet) during lactation period (11). The fourth group received lead acetate (10 mg /kg B.W. orally) concomitantly with vitamin C (100 mg /kg B.W.orally) during lactation period (12).

At the end of the experiment (21day) body weights of dams and their pups were recorded, blood samples collected from dams and their pups from the eye vein and placed in tubes which contain anticoagulant (EDTA) for complete blood picture. Also blood samples were collected into clean dry centrifuge tubes allowed to clot, serum separated after centrifugation at 1500 rpm for 15 minute. Hemoglobin concentration was determined by Sali method (13). Red and White blood cells count were counted per cubic millimeter of blood by Hemocytometer (13). Packed cell volume determined by microhematocrite capillary tube and Microhematocrite centrifuge (12000 circle / minute) for 10 minutes (13). Differential Leucocytes count (DLC) Thin blood film was prepared and stained with Gimza (13). Mean corpuscular volume (MCV), mean corpuscular Hemoglobin (MCH) and mean corpuscular Hemoglobin concentration (MCHC) were calculated (13). Determination of total protein by using Biuret method (14).

## Statistical analysis

Our data were analyzed statistically using one – way analysis of variance. Group differences were determined

using Duncan multiple range test. Statistical significance was considered at (P<0.05) (15).

**Results**

Administration of dams with lead acetate during the lactation period (20days) produced a significant decrease (P<0.05) in body weight, packed cells volume, hemoglobin concentration and red blood cells count as compared with the control group (Table 1), whereas a significant increase (P<0.05) in the white blood cells count and total proteins compared with control group. Treatment dams with vitamin E or C concomitantly with lead acetate during lactation period caused a significant increase (P<0.05) in packed cell volume compared with lead acetate group, and closed to control value group. Treatment with vitamin E or C did not affect significantly in body weight, red blood cells count, white blood cells count and total proteins compared with lead acetate group. Treatment with vitamin E and lead acetate caused a significant increase (P<0.05) in the hemoglobin concentration. But vitamin C did not affect significantly in hemoglobin concentration compared with group that treated with lead acetate alone.

Table (2) shows a significant increase (P<0.05) in monocytes percentage in lead acetate group compared with control group. No significant differences occurred between groups in the percentage of neutrophiles, lymphocytes, esinophiles and basophiles.

Treatment of dams with lead acetate caused a significant increase (P<0.05) in the mean corpuscular volume (MCV) and significant decrease (P<0.05) in mean corpuscular heamoglobin concentration (MCHC) (Table 3). Treatment with vitamin E and lead acetate produced a significant increase (P<0.05) in the mean corpuscular heamoglobin concentration (MCHC) as compared with lead acetate group. Treatment with vitamin C produced significant increase (P<0.05) in MCV as compared with lead acetate group.

Table (1) Body weight, packed cell volume, hemoglobin concentration, red blood cell count, white blood cell count and total proteins in lactating female rats treated with lead acetate and its interaction with vitamin E and C.

| Treatment groups | Body weight g | Packed cell volume % | Hemoglobin concentration g/100ml | Red blood cell count $\times 10^6$ | White blood cell $\times 10^3$ | Total Protein g/100ml |
|---|---|---|---|---|---|---|
| Control (Distal water orally) | 261.4±1.4 a | 31.8±0.5 a | 15.4±0.8 a | 5.6±0.2 a | 3.7±0.4 b | 4.38±0.7 b |
| Lead acetate (10mg/kg B.W. orally) | 198.4 ± 7.8 b | 28.2±0.7 b | 10.9±0.6 b | 3.4±0.3 bc | 5.4±0.4 a | 6.66±0.9 a |
| Lead acetate (10mg/kgB.W.orally) +Vitamin E (600mg/diet) | 229 ± 10.1 b | 34±1 a | 15.7±0.8 a | 4.1±0.1 b | 4.6±0.2 ab | 6.32±0.2 ab |
| Lead acetate (10mg/kgB.W.orally) +Vitamin C (100mg/kg B.W. orally) | 197.4± 16.1 b | 38±1.3 a | 11.5±0.7 b | 4.3±0.2 c | 4.7±0.3 ab | 7.13±0.6 a |

Values were expressed as means ± S. E., Values with different letters in column are significantly different (P<0.05), Number of animals 5 female rats / group.

Table (2) The percentage of neutrophils, lymphocytes, eosinophils, basophiles and monocytes in lactating female rats treated with lead acetate and its interaction with vitamin E or C.

| Treatment groups | Neutrophils % | Lymphocytes % | Eosinophils % | Basophiles % | Monocytes % |
|---|---|---|---|---|---|
| Control (Distal water orally) | 20.2±2.6 a | 75.2±3.1 a | 2±0.4 a | 1.4±0.2 a | 1.4±0.2 b |
| Lead acetate (10mg/kg B.W. orally) | 23.8±1 a | 70±0.5 a | 1.2±0.4 a | 1.8±0.2 a | 3.8±0.9 a |
| Lead acetate (10mg/kgB.W.orally) +Vitamin E (600mg/diet) | 21.6±1.1 a | 73.8±1 a | 1.4±0.5 a | 1.4±0.4 a | 1.4±0.4 b |
| Lead acetate (10mg/kgB.W.orally) +Vitamin C (100mg/kg B.W. orally) | 18.2±2 a | 77.6±2.3 a | 1±0.5 a | 1.2±0.4 a | 1.4±0.6 b |

Values were expressed as means ± S. E., Values with different letters in column are significantly different (P<0.05), Number of animals 5 female rats / group.

Table (3) Mean corpuscular volume, mean corpuscular heamoglobin and mean corpuscular hemoglobin concentration in lactating female rats treated with lead acetate and its interaction with vitamin E or C.

| Treatment groups | Mean corpuscular volume fl | Mean corpuscular Heamoglobin pg | Mean corpuscular Heamoglobin Concentration g/100ml |
|---|---|---|---|
| Control (Distal water orally) | 57.2±2.1 c | 27.6±1.3 b | 48.4±2.1 a |
| Lead acetate (10mg/kg B.W. orally) | 85.8±8.1 b | 33.4±3.8 ab | 38.6±1.8 b |
| Lead acetate (10mg/kgB.W.orally) +Vitamin E (600mg/diet) | 83.4±2.9 b | 38.6±2.9 a | 46.2±3.1 a |
| Lead acetate (10mg/kgB.W.orally) +Vitamin C (100mg/kg B.W. orally) | 103.8±4.1 a | 35.4±1.4 a | 33.8±1.3 b |

Values were expressed as means ± S. E., Values with different letters in column are significantly different (P<0.05), Number of animals 5 female rats / group.

Treatment of dams with lead acetate during the lactation period produced a significant decrease (P<0.05) in body weight, packed cell volume, hemoglobin concentration and red blood cell count accompanied with a significant increase in white blood cell count and total proteins in their pups compared with pups of control group (Table 4). Treatment of dams with vitamins E or C produced a significant increase (P<0.05) in the packed cell volume, hemoglobin concentration and red blood cell count of their pups as compared with the pups of the lead acetate group. Treatment of dams with vitamin E concomitantly with lead acetate produced a significant decrease (P<0.05) in white blood cell count of their pups.

Administration of dams lead acetate produced significant decrease (P<0.05) in the percentage of neutrophils accompanied with a significant increase (P<0.05) in the percentage of lymphocytes and monocytes in their pups compared with pups of control group (Table 5). Treatment the dams with vitamin E and lead acetate concomitantly produced significant increase (P<0.05) in the percentage of neutrophils, with significant decrease in the percentage of lymphocyte in their pups compared with lead acetate group.

Table (4) The body weight, packed cell volume, hemoglobin concentration, red blood cell count, white blood cell count and total proteins in pups from dams treated with lead acetate and its interaction with vitamin E or C.

| Treatment groups | Body weight g | Packed cell volume % | Hemoglobin concentration g/100ml | Red blood cell count $\times 10^6$ | White blood cell $\times 10^3$ | Total Protein g/100ml |
|---|---|---|---|---|---|---|
| Control (Distal water orally) | 25.2±0.3 a | 23.9±0.3 b | 9.5±0.3 a | 3.9±0.2 a | 3.5±0.1 b | 4.27±0.3 b |
| Lead acetate (10mg/kg B.W. orally) | 14.9±0.5 d | 21±0.9 c | 7.2±0.3 b | 2.5±0.1 c | 4.7±0.4 a | 6.85±0.5 a |
| Lead acetate (10mg/kgB.W.orally) +Vitamin E (600mg/diet) | 23±0.7 b | 26.9±0.7 a | 9.6±0.3 a | 3.1±0.1 b | 3.7±0.2 b | 5.9±0.3 a |
| Lead acetate (10mg/kgB.W.orally) +Vitamin C (100mg/kg B.W. orally) | 18.4±0.7 c | 26.8±0.4 a | 9±0.2 a | 3.1±0.2 b | 4.7±0.2 a | 6.52±0.5 a |

Values were expressed as means ± S. E., Values with different letters in column are significantly different (P<0.05), Number of animals 22 pups / group.

Mean corpuscular heamoglobin concentration decreased significantly (P<0.05) (Table 6), whereas the mean corpuscular volume significantly increased (P<0.05) in pups from lead acetate treated dams as compared with control group.

**Discussion**

The results of the present study demonstrated that lead acetate administration to female lactating rats during result in significant decrease in body weight, PCV, Hb, RBC count, MCHC and a significant increase in WBC count,

MCV, total proteins in the dams and their pups compared with control group. This finding therefore corroborates similar findings reported by Teijon *et al* (16), Mugahi *et al* (17) and Khan *et al* (18) in mice, whom observed a significant decrease in body weights, erythrocytes number, hemoglobin concentration in rats that injected with lead as acetate acetic acid. This decrease in the body weights can be due to gradual toxicity that lead causes in the animals (16). Lead acetate is a toxic metal known to exert oxidative stress on multiple organs (19), it can be transferred from the maternal to the infant through milk when there is maternal exposure to lead (3). Anemia is one of the early manifestations of lead poisoning, it result from reduction of the life span of circulating erythrocyte as well as by inhibition the body's ability to make heamoglobin by interfering with several enzymatic steps in hem pathway (1,16). Ferrochelatase, which catalyze the insertion of iron into protoporphyrin IX, is quite sensitive to lead (1). Erythrocyte Na-K-ATPase is some what inhibited by lead suggesting a loss of cell membrane integrity this may account for the shortened lifespan of erythrocytes (1), also lead can cause damage in the erythrocytes originating defective cells that are eliminated by spleen and their hemolysis (16).

Table (5) The percentage of neutrophils, lymphocytes, eosinophils, basophiles and monocytes in pups from dams treated with lead acetate and its interaction with vitamin E or C.

| Treatment groups | Neutrophils % | Lymphocytes % | Eosinophils % | Basophiles % | Monocytes % |
|---|---|---|---|---|---|
| Control (Distal water orally) | 23.9±1.7<br>a | 71.3±1.9<br>b | 1.8±0.3<br>a | 1.8±0.7<br>a | 1.4±0.4<br>b |
| Lead acetate<br>(10mg/kg B.W. orally) | 19.2±0.7<br>b | 75.8±0.7<br>a | 0.8±0.1<br>a | 2.4±0.2<br>a | 2.8±0.3<br>a |
| Lead acetate (10mg/kgB.W.orally)<br>+Vitamin E (600mg/diet) | 23.3±0.7<br>a | 70.5±1<br>b | 1.0±0.6<br>a | 1.3±0.2<br>a | 1.9±0.4<br>ab |
| Lead acetate (10mg/kgB.W.orally)<br>+Vitamin C (100mg/kg B.W. orally) | 18.4±0.7<br>b | 76.6±0.9<br>a | 0.2±0.1<br>b | 2.7±1.2<br>a | 2.1±0.5<br>ab |

Values were expressed as means ± S. E., Values with different letters in column are significantly different (P<0.05), Number of animals 22 pups / group.

Table (6) The mean corpuscular volume, mean corpuscular hemoglobin\ and mean corpuscular heamoglobin concentration in pups from dams treated with lead acetate and interaction with vitamin E or C.

| Treatment groups | Mean corpuscular volume<br>fl | Mean corpuscular Heamoglobin<br>pg | Mean corpuscular Heamoglobin Concentration<br>g/100ml |
|---|---|---|---|
| Control (Distal water orally) | 63.9±4.7<br>b | 28.1±3.5<br>a | 39.7±1.3<br>a |
| Lead acetate<br>(10mg/kg B.W. orally) | 86.1±2.5<br>a | 30.1±1.3<br>a | 34.5±1.7<br>b |
| Lead acetate (10mg/kgB.W.orally)<br>+Vitamin E (600mg/diet) | 90.2±2.8<br>a | 32.3±1.4<br>a | 35.8±1.6<br>b |
| Lead acetate (10mg/kgB.W.orally)<br>+Vitamin C (100mg/kg B.W. orally) | 87±5<br>a | 29.4±1.7<br>a | 34.1±1.1<br>b |

Values were expressed as means ± S. E., Values with different letters in column are significantly different (P<0.05), Number of animals 22 pups / group.

In the present study the increase in the mean corpuscular volume and the decrease in the mean corpuscular hemoglobin concentration of dams that treated with lead acetate and their pups may result from the toxic effect of lead acetate that affect on red blood cells count and heamoglobin concentration, because the validity of these indexes is influenced by the value of red cell count, hemoglobin concentration and packed cell volume (13).

In the present study oral administration of lead acetate caused significant increase in the white blood cell count and monocytes percentage in the dams and their pups, on the other hand treatment of dams with lead acetate produced significant increase in the lymphocyte percentage and significant decrease in the neutrophils percentage in their pups. Opposite results has been reported by other investigators (16), suggested no changes in the white blood

cell count in rats treated with lead. Gheng and others (20) Have reported that lead increases lipopolysacharide (LPS), lead to liver injury and over expression of monocytes and macrophages, lipopolysacharide causes liver injury at high doses but non injurious inflammation at low doses, this may result in the increase of white blood cell count. These different findings reported in the peripheral blood cell is probably result from effect of lead on progenitor cells (20).

Concomitant administration of vitamin E and lead acetate during the lactation produced significant increase in the packed cell volume, heamoglobin concentration, mean corpuscular heamoglobin concentration, and significant decrease in the monocytes percentage of dams. In addition treatment of the female rats with vitamin E and lead acetate produced significant increase in the packed cell volume, heamoglobin concentration, red blood cell count, neutrophils percentage, and significant decrease in white blood cell count, lymphocyte percentage of their pups. Similar results has been reported by (21), suggested that administration of vitamin E to dams exposed to oxidative stress by cadmium chloride during lactation decreased the adverse effects produced by cadmium chloride on blood parameters in weaned pups. The main function of vitamin E is as a chain – breaking free radical trapping antioxidant in cell membranes and plasma lipoproteins, it react with lipid peroxide radicals formed by peroxidation of polyunsaturated fatty acids before they can establish chain reactions (10), vitamin E play a role in the protecting tissue from oxidative damage especially at the early stages of life, it transported to the infant during the lactation and pregnancy (22). Murray and others (10) reported that deficiency of vitamin E produce anemia in the infant by hemolysis of red blood cells and decrease in the heamoglobin production. Vitamin E deficiency enhances the susceptibilities of animals to hemolytic effect of lead poisoning (23), vitamin E useful in order to protect membrane-lipids and to prevent protein oxidation produced by lead intoxication (24).

On the other hand vitamin C given with lead acetate in the group 4 produced significant increase in the packed cell volume in the dams, in the pups vitamin C treatment result in significant increase in the packed cell volume, heamoglobin concentration, red blood cell count. Recent studies by (21) has shown that treatment of dams with vitamin C and cadmium chloride during the lactation period produced significant increase in heamoglobin concentration, packed cell volume in the dams and their pups. Ascorbic acid is great antioxidant and helps in the protecting the body against pollutants (25). The effects of vitamin C on blood parameters and anemia may result by increasing the iron absorption in gastrointestinal tract (GIT) and enhances iron bioavailability (26). Wang and others (27) reported that vitamin C is effective in reversing reactive oxygen species (ROS) induced mouse embryo toxicity. As an antioxidant vitamin C protect folate, vitamin E and polyunsaturated substance from destruction by oxygen as they move throughout the body (28).On the other hand vitamin C did not reverse some of blood parameters like white blood cell count, total protein, MCV, MCHC in the dams and pups this may result from the dose of vitamin C, rout of administration, duration of treatment, individual variations between the animals.

## References


1. Doull J, Klaassen CD, Amdur MO. Casaratt and Doulls Toxicology. 2nd ed. United state, Macmillan publishing co. 1980. pp:415–421.
2. Ettinger AS, Tellez – Rojo MM, Amarasiriwardena C, Gonzalaz – Avila M. Levels of lead breast milk and their relation to maternal blood and bone lead levels at one mounth postpartum. Environmental health perspectives 2004;112 (8):926 –931.
3. Gulson BL, Jameson C W, Mahaffey KR, Mizon K J, Patison N, Law AJ, Korsch MJ, Salter MA. Relationships of lead in breast milk to lead in blood, urine, and diet of infant and mother. Environ Health Perspe. 1998;106 (10):667–674.
4. AL- Jobory STA. Reproductive efficiency of suckling rats treated with lead acetate during lactation:Role of vitamin E. M.SC. Thesis, College of Veterinary Medicine, University of Mosul, 2006; pp:50. (in Arabic).
5. Flora S, Pande M, Kannan GM, Mehta A. Lead induced oxidative stress and its recovery following co-administration of melatonin or n-acetylcysteine during chelation with succimer in male rats. Cell Mol Biol 2004;50:543–551.
6. Kang JK, Sul D, Kang JK, Nam S, Kim H, Lee E.Effect of lead exposure on the expression of phospholipids Hydroperoxidase Glutathione peroxidase mRNS in the rat brain. Toxicolo Sci. 2004;82 (1):228–236.
7. Vaziri ND, Liang K, Ding Y. Increased nitric oxide inactivation by reactive oxygen species in lead – induced hypertension. Kidney Int 1999;56 (4):1492–1498.
8. Ferrari CKB. Oxidative stress pathophysiology:Searching for an effective antioxidants protection. Int Med J 2001;8 (3):175–184.
9. Bendich A. Antioxidant micronutrients and immune response in Bendich and Chandra, R.K. (eds), Micronutreints and immune function, Annals New York Academy of Sciences 1990;587:172–178.
10. Murray RK, Granner DK, Mayes PA, Rodwell VW. Harpers illustrated biochemistry. 27th ed. United States of America, Mc Graw – Hill Companies. 2007;pp: 489–505.
11. Alkattan M. Effect of using some antioxidants on production performance and some physiological characters in laying hens. Ph.D dissertation, College of Agriculture and forestry, University of mosul,2006.28p.(in Arabic).
12. AL-Babily EOH. Effect of sodium nitrate on spermatogenesis in rats. M.Sc. thesis, College of Veterinary Medicine, University of Mosul. 2007; pp:22. (in Arabic).
13. Jain NC ed. Schalm s veterinary hematology. USA, Lea and Febiger 1986;pp:276–82.
14. Wotton LD. D. Micro analysis in medical biochemistry. 5th ed. Edinburgh, Churchill Livingstone 1974; pp:156–159.
15. Bruning JL, Kintz BL. Computation handbook of statistics. Illinois:Scott, Foresman and Co. 1977.
16. Teijon C, Delsocorro JM, Martin JA, Lozano M, Bernardo V, Blaco D. Lead accumulation in rats at non acute doses and short periods of time:Hepatic, renal and hematological effects. Ecotoxico Environm Restor. 2000;3(1):36 –41.
17. Mugahi MH. Heidari Z. Sagheb HM. Barbarestani M. Effect of chronic lead acetate intoxication on blood indices of male adult rats. Daru 2003:11(4) 147-151.

18. Khan MSH. Mostofa M. Jahan MS. Sayed MA. Hossain MA. Effect of garlic and vitamin B-complex in lead acetate induced toxicities in mice.Bangl. J Vet Med. 2008;6(2):203-210.
19. Hus PC, Guo YL. Antioxidant nutrients and lead toxicity. Toxicol.2002;180 (1):33–44.
20. Gheng Y, Yang B, Yie liu M. Lead increases lipopolysaccharide – induced liver – injury through tumor necrosis factor – α overexpression by monocytes / macrophages:Role of protein kinase C and P42/44 mitogen –activated protein kinase. Environmental Health Perspectives 2006;114 (4):507–513.
21. Hassan AA, Effect of antioxidants on certain physiological and biochemical aspects in female rats treated with cadmium during pregnancy and lactation. Ph.D thesis, College of veterinary Medicine, University of Mosul. 2006;PP:35p. (in Arabic).
22. Debier C, Larondelle Y. Vitamin A and E:metabolism, roles and transfer to offspring. Br J Nutr 2005;93(2):153 – 174.
23. Levander OA, Morris VC, Higgs DJ, Ferretti RJ. Lead poisoning in vitamin E-deficint rats. J Nutr 1975;105 (11):1481 –1485.
24. Rendon – Ramirez A, Cerbon – Solorzano J, Maldonado – Vega M, Quintanar – Escorza MA, Galderon – Salinas JV. Vitamin – E reduces the oxidative damage on delta – aminolevulinic dehydratase induced by lead intoxication in rat erytheocytes. Toxicol In vitro 2007;21 (6):1121–1126.
25. Zahang J, Jiang S, Watson RR, Antioxidant supplementation prevents oxidation and inflammatory responses induced by sidestream cigratte smoke in old mice. Environ Health Persp 2001;109:1007 – 1009.
26. Marisnn G, Shari H, Brenda M. The role of antioxidants in arsenic and cadmium toxicity. Altern Med Rev 2003;8 (2):106–128.
27. Wang X, Falcone T, Attaran M, Goldberg JM, Agarwal A, Sharma F. Vitamin C and vitamin E supplementation reduce oxidative stress induced embryo toxicity and improve the blastocyst development rate. Fertil Steril 2002;78 (6):1272 – 7.
28. Grodner M, Long S, De young S. Foundations and clinical applications of nutrition anursing approach. 3ed ed. China, Mosby 2004. PP: 167 – 199.