J.Agric.Chem.and Biotechn., Mansoura Univ.Vol. 8(7):191 – 196, 2017

Antioxidant Effect of Ascorbic Acid (Vit.C) in Different Concentration against Harmful Effect of Toxic Heavy Metal (Lead Acetate) in Rats

Lamia M. Hafez and Amal M. Kishk

Regional Center for Food and Feed - Agriculture Research Center- El-Sabhia, Alexandria-Egypt

ABSTRACT

Ascorbic acid (Vit.C) is one of the most potent antioxidants that interacts directly with the oxidizing radicals and protects the cells from reactive oxygen species. There is a need to increase the daily intake of Vit.C. The present study was aimed to evaluate the potential protective effects of different concentration of Vit.C against oxidative stress of lead acetate toxicity (Pb) in rats. Fifty albino male rats with average body weight 180 gm were classified into five equal groups (10 each); control group received tap water only, Pb group received 0.2% lead acetate /kg,b.w and the other three groups received 500, 1000 and 1500 mg Vit. C, along with 0.2%lead acetate /kg.bw, respectively. Doses were orally administered daily for two months.The results showed that Pb affected significantly on the hematological parameters, which decreased red blood cell count (RBC) by 28.9%, hemoglobin(HB) by 28.2%, hematocrit (Htc) by 26.4%and platelets(Plt) by 25,1% than control group ;while white blood cell count (WBC) increased significantly by 35.9% than control group. But Vit.C enhanced hematological parameters in the present of lead acetate at all concentrations than lead group, and the best effect was found with high concentration (1500mg),where it increased RBC count by 27.5%,Hb by 44.7%,Htc by 30.2%and Plt by 45%,while WBC count was decreased by 29%.Pb caused significant increase in total lipid by 53.7%, triglyceride (TG) by 43.6%, cholesterol (CHO) by 50%, low density lipoprotein (LDL)by 76% and very low density lipoprotein(VLDL) by 43.8% than control group, while Vit.C significantly decrease the total lipid by 26.1%, TG by 20.5%,CHO by 21.3%.LDL by 50%, and VLDL by 2o.5% at high concentration of Vit C(1500mg),it also increased HDL significantly by 57.5% than lead group. Pb administration increased nitric oxide (NO) by 53.9% and thiobarbaturic acid reactive substances(TBARS) by 53.3% in plasma compared to the control group, while it significantly decreased glutathione (GSH) by 50% and total antioxidants capacity (TAC) by 57.9%.Pb also increased NO and TBARS and decreased GSH and TAC in liver, kidneys and brain. Vit.C had benefits effect as antioxidant activity, the best effect noticed in the highest concentration of Vit.C (1500 mg).It caused significant decrease in NO by 23.4% and TBARS by 25.5%.But also caused significant increase in GSH by 60% and TAC by 100% in plasma than lead group. Vit C also decreased NO and TBARS and increased GSH and TAC in organs than lead groupIt can be concluded that Vit.C is capable of alleviating the harmful effects of Pb and highly recommended increasing the daily intake of Vit. C either from food rich in ascorbic acid or from supplementation.

Keywords: *Ascorbic acid, Lead, Oxidative stress, Lipid, Antioxidants, rats.*

INTRODUCTION

Ascorbic acid (Vit.C) is low molecular mass antioxidant that interact directly with the oxidizing radicals which was widely reported with the capability of protecting cells from oxidative stress (Patra and Swarp 2004) and protect the cells from reactive oxygen species (Rai *et al* 2009).It also has been shown to regenerate other antioxidants within the body, including vitamin E(Jacob and Sotoudeh 2002).

Much of the current focus on Vit.C has been on its role as an antioxidant. Antioxidant nutrients are thought to provide a buffer against cell damage that occurs during routine cell functions. Studies examine the eating habits of large groups of people show that a high intake of Vit.C-rich foods may reduce oxidative stress which conceder a reason for cancers of the mouth, pharynx, esophagus, stomach, lungs, and pancreas. Furthermore, Vit. C is widely reputed to have chemo preventive effects in doses greater than the current RDA of 60 mg/ day. High levels of AA are usually found in diets that contain more servings of fruits and vegetables are richer in other antioxidants. (Levine *et al* 1999)

Based on the observation that free radical was generated during the pathogenesis processes induced by Pb exposure, it was presumed that supplementation of antioxidants could be an alternative method for chelation therapy (Flora *et al* 2003).

Pb is a persistent metal that is still present in the environment (Garaza and Soto 2006). Exposure to Pb mainly occurs through the respiratory and gastrointestinal systems. Absorbed Pb (whether inhaled or ingested) is stored in soft tissues. Pb is conjugated in the liver and passed to the kidneys, where a small quantity is excreted in urine and the rest accumulates in various body organs, affecting many biological activities which may result in morphological alterations that can remain even after Pb levels have fallen (Sidhu and Nehru 2004,Taib *et al* 2004 and Flora *et al* 2006). Researches in humans and animals have shown that Pb has toxicological effect on the liver, bone, blood, kidneys, lungs, heart, brain and testis, (Diamond 2005,Aziz *et al* 2012,Samia El-Neweshy and Yasmeen El-sayed 2011,Payal *et al* 2009,Bomfim *et al* 2012,Garu and Sharma 2011and Moussa and Bashandy 2008)

Pb mediated toxicities have been reported to include the induction of oxidative stress in tissues and organs which can lead to oxidative damage in two separate but related pathways. The first pathway is the generation of ROS and the second pathway is the depletion of antioxidants reserve through the generation of reactive oxygen species. Pb is reported to in activate glutathione an antioxidant by binding to its sulfhydryl moiety. (Ercal *et al* 1996 , Sharma and Garu 2011 and Patrick 2006).

The aim of this study was to evaluate the potential antioxidant effect of different concentration of ascorbic acid against oxidative stress resulted from toxic heavy metal such as lead acetate in rats.

MATERIALS AND METHODS

Experimental animals:

Male albino rats (*n:* 50) averaging 180 g of b.w were obtained from the animal house of the Medical

Research Institute, Alexandria University, Egypt. The local committee approved the design of the experiments and the protocol follows the guidelines of the National Institutes of Health (NIH). Animals received human care, and had adequate stable diet and water *ad* libitum. Animals were acclimatized to the laboratory conditions for two weeks before being experimented

Experimental design:

After two weeks of acclimation, animals were classified into five equal groups' (ten rats in each). Control group received tap water only, Pb group received 0.2% lead acetate /kg, b.w and the other three groups received 500, 1000 and 1500 mg AA along with 0.2% lead acetate / kg-Bw, respectively.Doses were orally administered every day for two months.

Body weight and organs weight:

Body weight of rats was recorded in the beginning and at the end of the experimental period. Animals were sacrificed by decapitation, then liver, kidneys, and brain immediately removed and weighed. Relative organ weights were calculated as g/100 g BW.

Blood sample:

Collected blood samples from the sacrificed animals in two separate tubes, one of them contained heparin used for estimation of hematological parameters using Celttacauto analyzer. These parameters included the red blood cell count, platelet, hematocrit and hemoglobin. The other tube contain EDTA was used for other analyses. Plasma samples were obtained by centrifugation at 4000 rpm for 20 min., then samples were stored at –20 ºC until using for further analyses.

Biochemical parameters:

Analyzed plasma samples were for CHO and TG according to the methods described by Tietz (1995). HDL was determined according to the methods of Sugiuchi *et al* (1995). LDL was determined by the method of Pisani*et al*., (1995). VLDL was calculated automatically by Roche /Hitachi Cobas C systems (TG /5).

Antioxidant enzymes and free radicals

The amount of GSH was determined according to Habig *et al.* (1974). TBARS were measured by Tappel and Zalkin (1959) .TAC and the level of NO were assayed in plasma and organs homogenates according to the manual instruction of Biodiagnostic Kit, Egypt.

Statistical analysis:

Steel and Torrie (1981) data were analyzed. Statistical significance of the difference between values of the control and treated animals was calculated by F test with 5% significance level. Data were statistically analyzed by using Duncan's Multiple Range Test SAS, (1986).

Table (1) showed the body weight gain and relative weight for liver, kidneys and brain for different groups. It was found that there was no significant change in body weight in all groups .Pb caused significant (≤0.05) increase in liver and kidneys weight than control group but it didn't effect on brain weight. Presence of Vit.C could enhance relative weight of liverat 500 mgand 1000 mg but the highest concentration (1500mg) didn't have any effect. Relative weight of kidneys increased at 500mg of Vit.C and decreased at 1000mg and 1500mgVit.C but it still more than lead group ,it was noticed an increase in brain weight at 1000mg Vit.C.

Table 1. Changes in body weight gain (g) and relative weight of organs (g/100 g BW) in different experimental groups.

Parameters	Experimental groups				
	Control group	Lead group 0.2% Pb	0.2% Pb +500mgAA	0.2% Pb +1000mgAA	0.2% Pb +1500mgAA
Initial weight	178.5 ± 5. 3^{a}	183.0 ± 6. 7^{a}	181.7 ± 5.9^{a}	185.7 ± 7. 2^{a}	185.8 ± 9.4^{a}
Final weight	215.5 ± 6.9^{a}	215.4 ± 14.7^{a}	210.0 ± 11.0^{a}	207.1 ± 13.3^{a}	211.7 ± 14.7^{a}
Body weight gain	37.0 ± 6.4^{a}	32.4 ± 6.8^{a}	28.3 ± 7.1^{a}	21.4 ± 10.6^{a}	25.8 ± 10.1^{a}
Liver	2.6 ± 0.05^{b}	3.2 ± 0.1^{a}	3.1 ± 0.04^{a}	3.1 ± 0.06^{a}	3.2 ± 0.1^{a}
Kidneys	0.7 ± 0.01^{c}	0.9 ± 0.03^{b}	1.1 ± 0.0 5^{a}	1.0 ± 0.02^{ab}	1.0 ± 0.06^{ab}
Brain	0.7 ± 0.04^{a}	0.7 ± 0.05^{a}	0.7 ± 0.03^{a}	0.8± 0.05^{a}	0.6 ± 10.05^{a}

Results are expressed as mean ± SE; Means with different letters in the same row imply significant differences at p ≤0.05.

Pb affected significantly on hematological parameters (table 2), it caused significant decrease in RBC count by 28.9%,Hb by 28.2%, Htc by 26.4%and Plt count by 25.1% than control group, while WBC count increased by 35.6% than control group. Vit C enhanced hematological parameters in the present of lead acetate at all concentrations than lead group, and the best effect was found with the highest concentration (1500mg), where it increased RBC count by 27.5%,Hb by 44.7%,Htc by 30.2%and Plt count by 45%,while WBC count decreased by 29%.,which became near to control group .From the table it was noticed that there is no difference in Plt count in all concentration of Vit C in presence of lead.

Blood lipid was affected by lead acetate, Pb caused significant increase in total lipid by 53.7%, TG by 43.6%,CHO by 50%,LDL 76% and VLDL by 43.8% than control group .Pb also decreased significantly HDL than control nearly by half (36.5±1,9 in Pb group and 65.7±2.1 in control group), while AA significantly decreased total lipid by 26.1%, TG by 20.5%,CHO by 21.3%.LDL by 50%, and VLDL 2o.5% at high concentration (1500mg),it also increased HDL significantly by 57.5% in the presence of lead acetate than lead group . There was no different effect between the two concentrations of 500 mg and 1000 mg which caused nearly the same effect on lipids. (table3).

Antioxidant activity presented in table (4), it was found that Pb increased significantly the level of NO by 53.9% and TBARS by 53.3% in plasma than control group. Pb also caused significant decrease in the level of GSH by 50% and TAC by 57.9% in plasma than control group .Pb also increased NO and TBARS and decreased GSH and TAC in organs than control group, while all concentrations of Vit C had benefits effect on antioxidant activity, the best effect noticed in the highest concentration of Vit.C (1500 mg).It caused significant decrease in NO by 23.4% and TBARS by 25.5%.It also caused significant increase in GSH by 60% and TAC by 100% in plasma than lead group. Vit.C also decreased NO and TBARS and increased GSH and TAC in organs than lead group.

Table 2. Changes in hematological parameters in different experimental groups.

Parameters	Experimental groups				
	Control group	Lead group 0.2% Pb	0.2% Pb +500mgAA	0.2% Pb +1000mgAA	0.2% Pb +1500mgAA
RBCs count($x10^6$L)	4.7 ± 0.2^{a}	3.34 ± 0.2^{d}	4.18 ± 0.12^{b}	3.84 ± 0.2^{c}	4.26 ± 0.2^{b}
WBCs count($x10^3$L)	7860 ± 716^{b}	10680 ± 878.3^{a}	8440 ± 882.9^{b}	8040 ± 1092.1^{b}	7580 ± 879.1^{c}
Hb(gm/dl)	13.1 ± 0.5^{ab}	9.4 ± 0.6^{d}	11.9 ± 0.4^{c}	12.5 ± 0.3^{bc}	13.6 ± 0.4^{a}
Htc(%)	43.2 ± 1.7^{a}	31.8 ± 2.8^{b}	41.2 ± 1.3^{a}	41.8 ± 1.6^{a}	41.4 ± 1.4^{a}
Plt count($x10^9$L)	241.8 ± 19.4^{ab}	181.2 ± 16.4^{c}	223.6 ± 12.5^{b}	234.6 ± 23.5^{ab}	263.2 ± 18.8^{a}

Results are expressed as mean ± SE; Means with different letters in the same row imply significant differences at p ≤0.05.

Table 3.Changes in blood lipid fractions in different experimental groups

Parameters	Experimental groups				
	Control group	Lead group 0.2% Pb	0.2% Pb +500mgAA	0.2% Pb +1000mgAA	0.2% Pb +1500mgAA
T. Lipid(mg/dl)	472.8 ± 20.3^{d}	726.6 ± 22.5^{a}	604.2 ± 12.3^{b}	591 ± 16.7^{bc}	536.7 ± 30.8^{c}
TG(mg/dl)	129.0 ± 2.5^{d}	185.3 ± 5.2^{a}	165.2 ± 3.4^{b}	162.7 ± 7.8^{b}	147.3 ± 3.4^{c}
CHO(mg/dl)	116.5 ± 4.1^{e}	174.8 ± 6.2^{a}	151.5 ± 5.4^{b}	148.0 ± 5.2^{c}	137.5 ± 4.9^{d}
HDL(mg/dl)	65.7 ± 2.1^{a}	36.5 ± 1.9^{d}	48.6 ± 2.3^{c}	52.1 ± 1.1^{c}	57.5 ± 0.95^{b}
LDL(mg/dl)	26.9 ± 4.7^{d}	101.2 ± 5.1^{a}	69.8 ± 5.6^{b}	$63.3. \pm 4.7^{b}$	50.5 ± 5.0^{c}
VLDL(mg/dl)	25.8 ± 0.5^{d}	37.1 ± 1.0^{a}	33.1 ± 0.7^{b}	32.5 ± 1.6^{b}	29.5 ± 0.7^{c}

Results are expressed as mean ± SE; Means with different letters in the same row imply significant differences at p ≤0.05.

Table 4. Changes in antioxidant parameters in different experimental groups

Parameters		Experimental groups				
		Control group	Lead group 0.2% Pb	0.2% Pb +500mgAA	0.2% Pb +1000mgAA	0.2% Pb +1500mgAA
plasma	NO µmol/ml	51.4 ± 1.2^{c}	79.1 ± 1.9^{a}	65.2 ± 1.6^{b}	64.2 ± 1.6^{b}	60.6 ± 1.5^{c}
	TBARS nmol/ml	9.2 ± 0.5^{d}	14.1 ± 0.2^{a}	11.9 ± 0.4^{b}	11.7 ± 0.3^{b}	10.5 ± 0.3^{c}
	GSH µmol/ml	1.0 ± 0.03^{a}	0.5 ± 0.02^{d}	0.7 ± 0.02^{c}	0.7 ± 0.02^{c}	0.8 ± 0.02^{b}
	TAC mmol/ml	1.9 ± 0.05^{a}	0.8 ± 0.01^{d}	1.4 ± 0.02^{c}	1.4 ± 0.03^{c}	1.6 ± 0.01^{b}
liver	NO µmol/gtissue	11.7 ± 0.1^{e}	17.2 ± 0.2^{a}	15.0 ± 0.1^{b}	14.5 ± 0.1^{c}	13.4 ± 0.1^{d}
	TBARS nmol/g tissue	14.5 ± 0.1^{d}	21.2 ± 0.2^{a}	18.3 ± 0.2^{b}	17.8 ± 0.2^{b}	16.5 ± 0.1^{c}
	GSH µmol/g	5.9 ± 0.1^{a}	3.3 ± 0.04^{d}	4.3 ± 0.1^{c}	4.5 ± 0.1^{c}	5.1 ± 0.1^{b}
	TAC mmol/g tissue	1.6 ± 0.05^{a}	0.8 ± 0.1^{c}	1.2 ± 0.1^{b}	1.3 ± 0.07^{b}	1.4 ± 0.1^{b}
kidney	NO µmol/g tissue	5.2 ± 0.04^{d}	7.7 ± 0.1^{a}	6.6 ± 0.1^{b}	6.4 ± 0.1^{b}	6.1 ± 0.1^{c}
	TBARS nmol/g tissue	27.0 ± 0.7^{d}	41.9 ± 0.2^{a}	34.3 ± 0.2^{b}	33.4 ± 0.8^{b}	30.9 ± 0.4^{c}
	GSH µmol/g tissue	4.6 ± 0.1^{a}	2.5 ± 0.04^{e}	3.3 ± 0.1^{d}	3.5 ± 0.1^{c}	3.9 ± 0.1^{b}
	TAC mmol/g tissue	17.4 ± 0.2^{a}	7.3 ± 0.5^{d}	12.5 ± 0.3^{c}	13.1 ± 0.3^{c}	14.6 ± 0.3^{b}
brain	NO µmol/g tissue	14.8 ± 0.4^{d}	22.5 ± 0.6^{a}	18.9 ± 0.5^{b}	18.3 ± 0.5^{bc}	17.3 ± 0.5^{c}
	TBARS nmol/g tissue	31.2 ± 0.5^{d}	45.3 ± 0.5^{a}	39.4 ± 0.6^{b}	38.4 ± 0.6^{b}	35.6 ± 0.6^{c}
	GSH µmol/g tissue	6.4 ± 0.1^{a}	3.7 ± 0.01^{d}	5.0 ± 0.1^{c}	5.1 ± 0.1^{c}	5.6 ± 0.05^{b}
	TAC mmol/g tissue	16.8 ± 0.3^{a}	9.4 ± 0.1^{d}	12.9 ± 0.4^{c}	13.4 ± 0.3^{bc}	14.2 ± 0.4^{b}

Results are expressed as mean ± SE; Means with different letters in the same row imply significant differences at p ≤0.05

DISCUSSION

Vitamin C (ascorbic acid) was widely reported to have the ability to protect cells from oxidative stress. It is the most important free radical scavenger in extracellular fluids, trapping radicals in the aqueous phase and protecting biomembranes from peroxidative damage (Rai*et al* 2009).Animal studies have suggested an antagonistic effect of Vit C on Pb absorption and toxicity, alsoVit C may chelate Pb(Dawson *et al* 1999).In the present study it was found that presence of AA with lead acetate significantly decreased the harmful effect of Pb on hematological parameters with all doses , especially with the highest concentration (1500 mg). Pb ingestion resulted in a significant decrease of Hb in blood (Wang *et al* 2007). In a study of the effects of dietary AA levels on hematological parameters it was found a significantly higher Htc value with 1686mg AA kg diet compared to 32 and 423mg AA kg^{-1} diets, also WBC count with 1686mg AA kg diet was significantly higher than those fed on 32–423 mg AA kg diet.(Sahkar*et al* 2015)

In this study it was found that; Pb caused increase in CHO, TG,LDL and VLDL and decreased in HDL, while adding Vit C in the presence of Pb caused beneficial effect on lipid profile. In previous study a significant positive correlation was observed between blood lead and total cholesterol and between blood lead and LDL cholesterol. (Ademuyiwa*et al* 2005). Sharma 2012 in a study on battery workers found that, the TG levels were higher in the battery workers than control group , also it was suggested that Pb exposure increases cholesterol synthesis.

Previous study reported that 1 g Vit C inhibited the increase in LDL susceptibility to oxidation after exercise, preventing this acute pro-atherogenic effect. (Sanchez *et al* 1998), McRae (2008) in another study found that at least 500 mg/d of Vit C supplementation, for a minimum of 4 weeks feeding, can result in a significant decrease in serum LDL and TG concentrations. However, there was a non-significant elevation of serum HDL .It was found a significant decrease in TG, LDL was seen in the group supplemented with 1000 mg AA. The dose of 500 mg AA, however, did not produce any significant change in any of the parameters studied. (Afkhami and Ardekani 2007). Using AA supplementation can be considered as a useful harmless, economical and convenient prophylactic agent for Pb-exposed population (Shahrabi *et al* 2006).

TBARS are produced by lipid peroxidation and are considered as indicators of oxidative stress (Karthikeyan*et al* 2007).In present study it was found that lead acetate caused increase in TBARS and NO, and decrease in TAC and glutathione in both plasma and the organs (liver, kidneys and brain).Lead ingestion resulted in a significant decrease of GSH in blood and GSH level in liver cells (Wang *et al* 2007).

Frei *et al*., (1989) showed that Vit.C appears to be particularly important in limiting oxidative lipid damage in biological systems. Numerous studies have demonstrated that under many different types of oxidizing conditions, amino acid forms the first line of antioxidant defense and effectively protects the lipids in plasma and lipoproteins against detectable peroxidative damage. Upasani *et al*., (2001) reported that peroxidative damage increased the level of lipid peroxidation and altered the antioxidant defense system (Adanaylo and Oteiza, 1999). Although high dose amino acid decreased lipid peroxidation more than the low dose but the difference was not significant Shokouhi (2005).

CONCLUSION

From the present study, it was found that; the beneficial role of Vit.C is ameliorating lead-induced oxidative stress in lead exposed rats. Based on the observation that free radical was generated during the pathogenesis processes induced by lead exposure, it was presumed that supplementation with Vit.C was capable to protect cells from oxidative stress and help to improve the antioxidant status parameters by scavenging the generated free radicals, and highly recommended increasing the daily intake of Vit.C either from food rich in Vit C or from supplementation.

REFERENCES

Ademuyiwa, O.; R.N. Ugbaja, F. Idumebor and O. Adebawo.(2005).plasma lipid profiles and risk of cardiovascular disease in occupational lead exposure in Abeokuta, Nigeria., Lipids .Health Dis . 28:4-19.

Adonaylo,V.N.;P.I. Oteiza.(1999) Lead intoxication: antioxidant defenses and oxidative stress in rat brain. Toxicol. , 135: 77–85.

Afkhami, A. M. and A. S. Ardekani .(2007). Effect of vitamin C on blood glucose, serum lipids & serum insulin in type 2diabetes patients. Indian J .Med. Res. , 126: 471-474.

Aziz, F.M.; I.M.Mauloodand M.A.Chawsheen (2012) .Effects of melatonin,vitamin C and E alone or in combination on lead-induced injury in liver and kidney organs of rats. IOSR J. Pharm, 2(5): 13-18.

Bomfim, K. M. ; C. A. Garcia; F. P. Reis, J. A. Absorption levels and morphological features of fetal organs in wistar rats treated with lead acetate.Int.J.Morphol., 30(1): 119-124.

Dawson, E. B. ;D. R. Evans; W. A. Harris ; M. C.TeterandW. J. McGanity(1999) The effect of ascorbic acid supplementation on the blood lead levels of smokers. J. Am .Coll. Nutr;18(2):166–70.Diamond, G.L.(2005) Risk Assessment of Nephrotoxic Metals. In: Tarloff, J. and L. Lash (Eds.), the Toxicology of the Kidney.CRC Press, London , pp: 1099-1132.

Ercal, N.,P.Treratphan;T.C. Hammond ; R.H. Mathews ; N.H. Grannemann and D.R. Spitz (1996). In vivo indices of oxidative stress in lead-exposed C57BL/6 mice are reduced by treatment with meso- 2, 3-dimercaptosuccinic acid or N-acetyl cysteine. Free Rad. Biol. Med ;21, 157–161.

Flora, S. J.; G. J. Flora and G. Saxena(2006) Environmental Occurrence, Health Effects and Management of Lead Poisoning. In: Casas, S.B. and J. Sordo (Eds.), Lead: Chemistry, AnalyticalAspects, Environmental Impact and Health Effects.Elsevier, The Netherlands, pp: 158-228.

Flora, S., M.PandeandA.Mehta.(2003) Beneficial effect of combined administration of some naturally occurring antioxidants (vitamins) and thiolchelators in the treatment of chronic lead intoxication. Chem Biol Interact;145:267-280.

Frei, B.,L. England and B.N. Ames .(1989)Ascorbate is an outstanding antioxidant in human blood plasma. Proc. Natl. Acad. Sci. U.S.A.; 86: 6377- 6381.

GarazaA,V. R. and E. Soto (2006). Cellular mechanisms of lead neurotoxicity. Med. Sci. Monitor, 12(3): 57-65.

Garu,U.,R. Sharma and I. Barber. (2011)Effect of lead toxicity on developing testis of mice. Int. J. Pharma. Sci. Res, 2(9): 2403-2407.

Habig, W. H. ;M. J. Pabst and W. B. Jakoby (1974) :Glutathione S-transferases. The first enzymatic step in mercapturic acid formation.The J.of Biol. Chemistry; 249: 7130-7139.

Jacob, R.A. and G. Sotoudeh(2002) . Vitamin C function and status in disease. NutrClinCare ;5:66-74.

Karthikeyan, K. ; B. B. R. Sarala, and D. S. Niranj ali. (2007). Cardioprotective effect of grape seed proanthocyanidins on isoproterenol-induced myocardial injury in rats. Int. J. Cardiol, 115: 326-333.

Levine, M. S. C. Rumsey , R. Daruwala, J. B. Park and Y. Wang. (1999) Criteria and recommendations for vitamin C intake. Nutr. Rev.; 57 (7) : 222-224.

McRae, M.P. (2008) Vitamin C supplementation lowers serum low-density lipoprotein cholesterol and triglycerides: a meta-analysis of 13 randomized controlled trials J Chiropr Med Jun; 7(2): 48–58.

Moussa,S.A.and S.A. Bashandy.(2008) Biophysical and biochemical changes in the blood of rats exposed to lead toxicity,Rom.J.Biophys;18(2): 123-133.

Patra,R.,and D. Swarup.(2004). Effect of antioxidant ascorbic acid,L-methionine on tocopherol alone or along with chelator on cardiac tissue of lead-treated rats. Veterinarski Arch;74: 235–44.

Patrick, L.(2006) Lead toxicity, a review of the literature. Part I: Exposure, evaluation and treatment.Altern Med Rev ;11(1):2–22.

Payal, B.,H.P. Kaurand.V. Rai .(2009) New insight into the effects of lead modulation on antioxidant defense mechanism and traceelement concentration in rat bone .InterdiscipToxicol.,2(1):18-23.

Pisani, T.,C.Gebski and E Leary. (1995) Accurate Direct Determination of Low Density Lipoprotein Cholesterol Using an Immuno separation Reagent and Enzymatic Cholesterol Assay.Arch.Pathol.Lab.Med ;119(12):1127-1135.

Rai, D.K., P.K. Rai,S.I. Rizvi,G.Watal and B. Sharma . (2009):Carbofuran-induced toxicity in rats: protective role of vitamin C.Exp.Toxicol.Pathol. ;61(6):531–5.

Sahkar E,H.Yun ,D.J.Kim ,S.K.kim, B.I. Lee and S.C.Bai.(2015):Effects of dietary vitamin C levels on tissue ascorbic acid concentration hematology, non-specific immune response and gonad histology, in broodstock Japanese , Anguilla japonica.Aquaculture;438(1):115-121.

Samia M. El-Neweshy and YasmeenS,El-Sayed.(2011): Influence of vitamin C supplementation on lead-induced histopathological alterations in male rats. Exp. Toxicol. Pathol, 63(3): 221-227.

Sánchez J,O. LQuesada ,A.Jorba , C.Payés , R. Otal , F. Serra-Grim and González-Sastre(1998). Ascorbic acid inhibits the increase in low-density lipoprotein (LDL) susceptibility to oxidation and the proportion of electronegative LDL induced by intense aerobic exercise., J Ordonez-Llanos Coronary artery disease ; 9(5): 249-55.

SAS (1986): Statistical Analysis System. User's Guide: Statistics,version 5 Edition SAS Inst., Inc., Cary, NC, U.S.

Shahrabi F;A.R.;I.M.Jalal.;H. SadrzadehYeganehand A.S.Farvidm.(2006): Effect Of 2-Week Ascorbic Acid Supplementation On Plasma Lead Levels InWorkers Occupationally Exposed To Lead.Journal of Rafsanjan University of Medical Sciences and Health Services,Summer ; 5 (2):130-137.

Sharma R. and U. Garu.(2011):Effects of lead toxicity on developing testes in swiss mice. Univ., J. Environ. Res. Technol;1(4): 390-398.

Sharma S.V,P. Kumar, V. Atam,A. Vermaand R.C. Murthy(2012): Original Research Paper Lipid Profiles with Increase Blood Lead Level: Risk of Cardiovascular Disease in Battery Workers of Lucknow City J Indian Acad Forensic Med. October-December,Vol.34(4) 328:0971-0973.

Shokouhi GS,A. Hadidchi,M. Ghorbanihaghjo, S.Rahbani-Noubar ,M.H. Panahi, Forouzanfar, et al.(2005):Neuroprotective Effect of Ascorbic Acid In Experimental Blunt Sciatic Nerve Injury n Rats. The internet J. Nutr. and Wellness; 1 (2):140-147

Sidhu, P. and B. Nehru .(2004): Lead intoxication: Histological and oxidativedamage in rat cerebrum and cerebellum.J. Trace Elem. Exp. Med; 17(1): 45-53.

Steel R. and J. Torrie (1981): Principle and Procedure of Statistics. A Biometrical Approach, 2nd edition. McGvaus Hill Booh Company, New York, USA.

Sugiuchi H, H. Okabe and T. Irie (1995):Direct Measurement of High Density Lipoprotein Cholesterol in Serum with Polyethylene Glycol Modified Enzymes and Sulfated Cyclodextrin.Clin.Chem; 41:717-723.

Taib N.T.,B.M. Jarrar and M.Mubarak.(2004): Ultrastructural alterations in hepatic tissues of white rats (Rattus norvegicus) induced by lead experimental toxicity. Saudi J. Biol. Sci.; 11(1): 11-20.

Tappel A.L. and H. Zalkin.(1959): Inhibition of lipidperoxidation in mitochondria by vitamin E. Arch. Biochem. and Biophys.;80:333-336.

Tietz N,(1995): Clinical Guid to Laboratory Tests, 3rdedition .Philadelphia PA: WB Saunders Company. 1995 130-131.

Upasani C, A.Khera and R.Balaraman (2001): Effect of lead with Vitamins E, C, or spirulina on malondialdehyde : conjugated dienes and hydroperoxides in rats. Indian J Exp. Biol. 2001; 39:70-74.

Wang C, J. Liang, C. Zhang, Y.X.Shi and Shi (2007): Effect of Ascorbic Acid and Thiamine Supplementation at Different Concentrations on Lead Toxicity in Liveri Ann. Occup. Hyg. 2007:Vol. 51(6):563–569.

التأثيرالمضاد للاكسدة للتركيزات المختلفه من حمض الاسكوربيك (فيتامين ج) ضد التأثيرالضار للمعادن الثقيله السامة (خلات الرصاص) في الجرزان

لمياء محمد حافظ وأمال محمد كشك

المركز الإقليمي للأغذية والأعلاف - مركز البحوث الزراعية - الصبحية، الإسكندرية – مصر

حمض الاسكوربيك (فيتامين ج)هوواحد من أكثر المواد المضادة للاكسدة القوية التي تتفاعل مباشرة مع الشوارد المؤكسدة ويحمي الخلايا من الأنواع المتفاعله مع الأكسجين. وقد وجد ان هناك حاجة لزيادة المتناول اليومي من فيتامين ج. تهدف الدراسة لتقييم التأثيرات الوقائية المحتملة لتركيزات مختلفة من فيتامين ج ضد الجهد التاكسدي الناتج عن خلات الرصاص في الجرزان. وقد تم تقسيم خمسين من ذكورجرزان الالبينو بمتوسط وزن للجسم ١٨٠ جم إلى خمس مجموعات متساوية (١٠ لكل مجموعه) .المجموعه الضابطه (تلقت ماء الصنبور فقط)؛ الثانيه (تلقت ٠.٢٪ خلات الرصاص /كجم من وزن الجسم) والمجموعات الثلاث الأخرى (تلقت ٥٠٠ و ١٠٠٠ و ١٥٠٠ ملجم حامض اسكوربيك جنبا إلى جنب مع ٠.٢٪ خلات الرصاص /كجم من وزن الجسم)، على التوالي. كانت الجرعات تجرع بالفم كل يوم لمدة شهرين.أظهرت النتائج أن الرصاص يؤثر بشكل معنوي علي خصا ئص الدم، فقد انخفضت خلايا الدم الحمراء بنسبة ٢٨.٩٪، الهيموجلوبين بنسبة ٢٨.٢٪، هيماتوكريت بنسبة ٢٦.٤٪ والصفائح الدمويه بنسبة ٢٥،١٪ مقارنه بالمجموعة الضابطه. في حين زادت خلايا الدم البيضاء بنسبة ٣٥.٩٪ مقارنه بالمجموعة الضابطة ولكن اضافه حمض الاسكوربيك حسن من خصائص الدم في وجود خلات الرصاص بكافة الجرعات مقارنه بمجموعة الرصاص، وكان أفضل تأثير وجد مع التركيز الاعلي من فيتامين ج (١٥٠٠ ملجم) حيث ارتفعت خلايا الدم الحمراء بنسبه ٢٧,٥٪ والهيموجلوبين بنسبه ٤٤,٧٪ والهيماتوكريت بنسبه ٣٠,٢٪ والصفائح الدمويه بحوالي ٤٥٪، بينما انخفضت كرات الدم البيضاء بنسبة ٢٩٪.مقارنه بمجموعه الرصاص. وقد تسبب الرصاص في زيادة معنوية في الدهون الكليه بنسبة ٥٣.٧٪ والدهون الثلاثية بنسبة ٤٣.٦٪ والكولسترول بنسبة ٥٠٪، الليبوبروتينات منخفضه الكثافة بنسبة ٧٦٪ والليبوبروتينات منخفضه الكثافة جدا بنسبة ٤٣.٨٪ مقارنه بالمجموعة الضابطه، في حين وجد أن جمض الاسكوربيك يقلل بشكل كبير من الدهون الكلية بنسبة ٢٦.١٪، الدهون الثلاثيه بنسبة ٢٠.٥٪، الكوليسترول بنسبة ٢١.٣ ٪ .الليبوبروتينات منخفضه الكثافه بنسبة ٥٠٪، والليبوبروتينات منخفضه الكثافه جدا بنسبه ٢٠,٥٪ وذلك مع اعلى تركيز من جمض الاسكوربيك (١٥٠٠ملجم)، كما زادت الليبوبروتينات الثقيله بشكل ملحوظ بنسبة ٥٧.٥٪ عن مجموعة الرصاص. وقد زاد الرصاص من مستوى أكسيد النيتريك بنسبة ٥٣.٩٪ والمواد التفاعلية للحامض الثيوباربتيوريك بنسبة ٥٣.٣٪ في البلازما مقارنة مع المجموعة الضابطة، في حين انخفض الجلوتاثيون بنسبة ٥٠٪ ومضادات الأكسدة الكليه بنسبة ٥٧.٩٪. (مستوى معنوية <٠,٠٥).و كان لحمض الاسكوربيك تأثير على النشاط المضاد للأكسدة، و أفضل تأثير لوحظ مع الجرعات العالية من حمض الاسكوربيك (١٥٠٠ ملجم)، حيث أدى إلى انخفاض معنوي في مستوي اكسيد النيتريك بنسبة ٢٣.٤٪ و حامض الثيوباربتيوريك بنسبة ٢٥.٥٪. كما تسبب زيادة كبيرة في الجلوتاثيون بنسبة ٦٠٪ و مضادات الاكسده الكليه بنسبة ١٠٠٪ في البلازما مقارنه بمجموعة الرصاص. كما ادي وجود حامض الاسكوربيك الي انخفاض اكسيد النيتريك وحامض الثيوباربتيوريك وزيادة الجلوتاثيون ومضادات الاكسده الكليه في الأعضاء المختلفه مقارنه بمجموعة الرصاص . ويوصي بشدة زيادة الاستهلاك اليومي من حمض الاسكوربيك (فيتامين ج) إما من طريق الغذاء المتناول ذو المصدر العالي من حمض الاسكوربيك أو من المكملات الغذائية .