Amelioration of aluminium - intake oxidative stress by some antioxidants in male albino rats

Ahkam M. El-Gendy
Zoology Dep. Faculty of science (Girls' branch), Al-Azhar University, Cairo, Egypt.

Abstract

Back ground:

Aluminum is potentially toxic to humans. The Agency for Toxics Substances and Disease Registry (ATSDR) reported that aluminum accumulates mainly in the bone, liver, testes, kidneys and brain. The goal of the present study was to assess in rats the pro-oxidant effects induced by Al^{3+} exposure, as well as the protective role of exogenous melatonin (M), vitamin E (vit. E) or *N*-acetylcystiene (NAC). The effect of aluminium (Al) alone or combined with antioxidants (M), (vit. E) or (NAC) on some physiological parameters and antioxidants in male albino rats were studied.

Material and methods:

The animals were assigned to 5 groups: control (group I); Al^{3+}–intake (53.5 mg $AlCl_3$/litre drinking water , group II) ; 5 mg melatonin/kg b.wt. plus $AlCl_3$ (group III); , or vitamin E(100 mg/kg b.w.) plus $AlCl_3$ (group IV)or 100mg *N*-acetylcystien plus $AlCl_3$ (group V). Rats were orally administered their respective doses daily for 30 days. At the end of the treatment period, blood was obtained. Thereafter, brain, liver, kidney and testes were removed. These tissues were processed to examine oxidative stress markers: reduced glutathione (GSH), superoxide dismutase (SOD), glutathione peroxidase (GSHpx) and lipid peroxidation end products {malondialdhyde(MDA) + 4- hydroxynonenal (4- HNE)}. Samples of these tissues were also used to determine Al^{3+} concentrations.

Results :

In Al- toxicated group ,serum glucose and total cholesterol levels, liver enzyme activities (ASAT and ALAT), as well as, lipid peroxidation end products {malondialdhyde (MDA) + 4- hydroxynonenal (4-HNE)} were elevated significantly in the brain , liver ,kidney and testes tissues when compared with control group. On the other hand, serum triglycerides and tissue (liver, kidney and testes) intracellular antioxidants glutathione (GSH) and superoxide dismutase (SOD) and liver glutathione peroxidase (GSHpx) activity decreased significantly. Brain GSH also decreased but SOD showed no significant changes. Melatonin, vit. E and NAC improved the levels of the different changed parameters when combined with Al. The most improved correction was recorded when Al^{3+} combined with vit. E followed by M ,then NAC. Serum Al^{3+} levels were increased in Al^{3+} treated group as well as groups exposed to Al^{3+} combined with vit. E, M or NAC when compared with control group. Al^{3+} could not be detected in tissues by atomic spectrophotometer (aluminium metal concentrations were below the limit of detection by AAS).

Conclusion:

The results show that Al^{3+} exposure promotes oxidative stress in different tissues while melatonin, vitamin E and N-acetylcystiene exert antioxidant actions in Al^{3+}-treated animals. The protective effects of these antioxidants against cellular damage caused by Al^{3+}-induced oxidative stress, together with its low toxicity, make them worthy of investigation as potential supplements to be included in the treatment of neurological disorders in which the oxidative effects must be minimized as well as protection against liver, kidney and testes damage by Al- exposure.
Dietary vitamin E supplementation may offer further protection.

Key words: Aluminium, melatonin, vitamin E, N-acetylcystiene, antioxidants, lipid peroxidation , MDA.

Introduction

Aluminum (Al) is the third most abundant element (8%) in the earth's crust and its compounds are distributed widely in nature (WHO 2009). It constituents of all soils, plants and animals (Yokel and McNamara, 2001 and Krewski *et al*.,2006). Although Al^{3+} is present in trace amounts in the biological material, it does not appear to be essential element (Goyer, 2004) and usually considered to have harmful effects on general health. In addition to occurring naturally in food and water , Al^{3+} is added to drinking water, many processed foods, cosmetics, toothpaste, antiperspirants and adjuvants in various parenteral preparations and pharmaceutical agents(Becaria *et al*., 2002 and Pournourmohammadi, *et. al*., 2008). Al^{3+} metal is used widely in different fields (cans, utensils, containers, automobile bodies, pigments,…). Al^{3+} hydroxide is used as antacid and has been used in the past to reduce phosphate accumulation in uremia (Yokel , 2000).

Aluminum is potentially toxic to humans. The Agency for Toxic Substances and Disease Registry (ATSDR) reported that aluminum accumulates mainly in the bone, liver, testes, kidneys and brain (ATSDR ,1990). Exposure to Al^{3+} could occur through three principal routs: 1) Inhalation of air contaminated with Al^{3+} compounds. 2) Oral ingestion of Al^{3+} dusts or with food and drinking water (WHO ,2009*)*. The ingestion pathway is the most significant route of transfer of Al^{3+} from the environment to animals and humans, and 3) Dermal rout (Akyol *et al.*,2004). In industrial settings, inhalation is the most important rout of Al entry into the body. This leads to absorption of Al into the blood with possible systemic intoxication (Polizzi, 2002). Gastrointestinal absorption is minimal, although accumulation and toxicity were observed after intake of high doses of Al^{3+} in persons with chronic renal failure (Arnich *et al*., 2004 and Stella *et al*.,2005). The richest natural dietary sources of Al^{3+} are herbs and tea leaves (Jansen *et al*. 2002). The consumption of foods containing aluminum-containing food additives are a major sources of aluminum in the diet (Saiyed and Yokel, 2005 and Soni *et al*. , 2001).

Aluminum is a neurotoxicant (Sood *et al*., 2011). Esparza *et al*.(2003) showed that aluminum exposure promoted oxidative stress in different neural areas of the animals, including those in which aluminum concentrations were not significantly increased. It has been shown to play a role in the etiology of uremia – and dialysis – associated disorders of the brain(dialysis encephalopathy) and bone Al^{3+} associated bone disease. Al^{3+} also has been proposed as an environmental factor that may contribute to some neurodegenerative diseases, including Alzheimer's disease (AD) .

It seems that Al^{3+} has varying effects on different organs associated with different exposure routs. However, Al^{3+} has a catalytic activity that produces free radicals stimulating oxidative injury in the brain (Christen, 2000 and Lemire *et al*., 2011).

In a review article, Mohammadirad and Abdollahi (2011) recorded a significant increase in LPO and inhibition of antioxidant enzymes by Al^{3+} in plasma (Ranjbar *et al*., 2008) , brain (Sood *et al*., 2011), testes (Yousef and Salama, 2009 and Khattab *et al*., 2010), kidney, renal cortex, serum, erythrocyte(Farina *et al.,* 2005) , hepatocyte, liver (Mailloux *et al*., 2011).

Hypothetically, since oxidative stress plays a pathogenic role in Al^{3+} toxicity, supplementation with antioxidants should attenuate oxidative stress and improve oxidative stress-mediated damage in Al^{3+} toxicity. Therefore , there is an urgent need to identify effective antioxidants with therapeutic potential to ameliorate Al^{3+} toxicity .

Melatonin (N-acetyl-5-methoxytryptamine) is the major product of the pineal gland in vertebrates. It is a well-known antioxidant and free radical scavenger. Moreover, its solubility in lipid and aqueous media, which allows it to cross morphophysiological barriers and enter subcellular compartments, permit melatonin to function as a highly effective inhibitor of oxidative damage(Esposito and Cuzzocrea, 2010) . It is a very potent and efficient endogenous free radical scavenger. It reacts with the highly toxic hydroxide radical and provide on – site protection against oxidative damage to different biomolecules (Reiter, 2000), It is also involved in the regulation of electron transfer, detoxifying reactive radical intermediates and control pre-oxidative processes (Tan *et al*,.2000).

Vitamin E, a lipid-soluble vitamin with antioxidant properties has an important role in protecting biological systems (Paulis *et al*., 2011). Vitamin E has a high antioxidant capacity and plays a fundamental biologic role, especially in protecting cells and tissues from

oxidative damage and prevents the formation of toxic oxidation products such as those formed from unsaturated fatty acids (Quiles *et al*., 2002 and 2006). Also, vitamin E is effective in scavenging lipid radicals (ROO-) and is recognized as a potent chain – breaking antioxidant, with the particular function of preventing lipid peroxidation in the membrane and lipoproteins (Lorenzoni and Ruiz-Feria, 2006).

N-acetylcysteine (NAC), a cysteine prodrug, has shown promise in numerous pathological conditions involving oxidative stress((Vosters and Neve, 2002 and Kamboj *et al*.,2006a). As a sulphydryl donor NAC contributes to the regeneration of glutathione and by directly acting as a free radical scavenger (Aydin *et al*., 2002). Various studies have shown that NAC administration has a beneficial effect against oxidative stress in neurodegenerative diseases (Pocernich *et al*., 2001 and Kamboj *et al.*, 2008). Prakash and Kumar (2009) suggested that N-acetyl cysteine has a neuroprotective effect against aluminium-induced cognitive dysfunction and oxidative damage in rats.

So, the objective of this work was to investigate the ability of (M), vit. E or NAC to resist oxidative damage on the rat brain, liver, kidney and testes during exposure to Al^{3+}. Also the study aimed to compare the protective effects of these antioxidants.

Material and methods

Animals and treatment:

The chosen dose of Al^{3+} was depended on the U.S. EPA survey of water supplies throughout the U.S., the maximum aluminum concentration reported in finished water where an aluminum compound was used as a coagulant was 5.35 mg/L(ATSDR, 1990) and multiplied by 10 i. e. the used dose was 53.5mg/ L drinking water (this limit is expected in developing countries).

Thirty male albino rats (180 – 200 gm) were used in this experiment.

They were randomized and housed six to a cage in Stainless steel cages containing sawdust bedding. They received standard rat chow and water *ad libitum*. The room conditions were maintained at 22±2 °C and 12/12-h light/dark cycle. The animals were divided into five groups each consists of 6 animals:

1- Control group without any treatment.
2- Aluminium- intake group (53.5 mg $AlCl_3$/litre drinking water).
3- Aluminium- intake group (53.5 mg $AlCl_3$/litre drinking water) and supplemented with 100mg vitamin E / kg b.wt. by gastric tube.
4- Aluminium- intake group(53.5 mg $AlCl_3$/litre drinking water) and supplemented with a daily dose of melatonin (M) 5 mg / kg b.wt. by using gastric tube.
5- Aluminium- intake group (53.5 mg $AlCl_3$/litre drinking water, and supplemented with NAC (100mg/kg b. wt.) in distilled water by gastric tube.

Chemicals:

All chemicals, Aluminium chloride ($AlCl_3$), Melatonin and N-acetyl cysteine Were purchased from Sigma Co. USA. Vitamin E "α-tocopherol acetate" capsules, supplied by Pharco Pharmaceutical Co., Egypt (each capsule contains 100 mg vitamin E).

After the experimentation period, the animals were fasted for 12 hours, and then sacrificed by sharp razor through jugular vein. The blood was collected; serum was separated and used for different analysis. The collected tissues (brain, liver, kidney and testes) of each animal were removed quickly, dried by filter paper, weighed and homogenized and kept at -20ºC for analysis.

Methods:

The concentrations of glucose and total cholesterol in serum were estimated by kits obtained from Stanbio, Texas, USA according to Tietz (1995) and triglycerides was measured by the method of Bucolo and David (1973). ASAT and ALAT activities in sera samples were estimated according to the method of Schumann *et al*.(2002) . GSH contents, SOD activity in the tested organs (brain, liver, kidney and testes) were determined by the methods of Prince and loose (1969) and Nishikimi *et al*. (1972) respectively. Hepatic GSHpx was measured according to the method of Ammerman, *et. al*. (1980) and Lipid peroxidation in the different tissues was estimated by colorimetric assay of malondialdhyde (MDA) + 4- hydroxynonenal (4- HDNE) as described by Esterbauer *et. al*. (1991), using kits from Wak- Chem Medical GMBH, Germany.

Al^{3+} was determined in all samples using Atomic Absorption Spectrophotometer (AAS), at AAS unit, Chemistry department, Faculty of Science, Mansoura University.

Data are expressed as mean ±S.D. Statistical analysis of the results was performed by ANOVA(SPSS program) followed by Post Hoc tests. A difference was considered significant when $p \leq 0.05$.

Results

The effect of Al^{3+} alone or combined with different antioxidants on serum glucose ,total cholesterol, and triglycerides levels as well as ASAT and ALAT activities are presented in table (1). The obtained data revealed significant increases in glucose , total cholesterol levels and ASAT and ALAT activities in Al^{3+}- exposed group when compared with control group, while serum triglycerides were decreased.Different antioxidants (vit. E, M and NAC) combined with Al^{3+} corrected these changes to nearly the control level.

Serum Al^{3+} concentrations are represented in table (1): It elevated in all Al^{3+}- exposed rats alone or combined with antioxidants. Atomic absorption spectrophotometer could not detect Al^{3+} in the different tested tissues (aluminium metal measurements were below the limit of detection by AAS).

Table (1): some serum parameters in Al^{3+}- treated rats and antioxidants (M, vit.E or NAC).

Parameters / Groups	control	Al^{3+}	Al^{3+}+ M	Al^{3+}+ vit. E	Al^{3+} + NAC
Glucose(mg/100ml)	77.038±0.928	86.558±7.532[a]	74.998±5.467[b]	78.963±7.143	77.460±6.086
Total cholesterol (mg/100ml)	92.985±12.597	105.938±19.799a	92.813±13.025	93.112±11.628	93.530±9.203
Triglycerides (mg/100ml)	56.980±4.696	47.643±2.022[a]	54.497±3.469[b]	56.387±3.653[b]	56.985±4.698[b]
ASAT (U/liter)	51.730±10.509	143.627±24.626[a]	57.503±6.226[b]	54.188±5.920[b]	51.730±10.509[b]
ALAT (U/liter)	40.807±6.546	63.592±7.695[a]	46.720±5.997[b]	43.213±6.607[b]	45.922±6.250[b]
Aluminium (Pg/liter)	0.232±0.066	8.443±1.219[a]	7.232±0.814[a]	8.332±1.517[a]	7.172±.686[a]

Mean ± SD of 6 animals in each group.
a Significant at P≤0.05 when compared with control group.
b Significant at P≤0.05 when compared with Al^{3+}- intake group.

In table (2): Brain GSH, SOD and {malondialdhyde(MDA)+4- hydroxynonenal (4-HNE)}in different groups are represented. Brain GSH decreased significantly and SOD insignificantly while (MDA)+4- HNE elevated significantly.

Table (2): some antioxidant parameters and {malondialdhyde(MDA) + 4- hydroxynonenal (4- HNE)}in the brain of Al^{3+}- treated rats and antioxidants (M, vit.E or ANAC).

Parameters / Groups	Control	Al^{3+}	Al^{3+}+ M	Al^{3+}+ vit E	Al^{3+}+NAC
GSH (µmol/g)	7.15± 0.49	5.15± 0.52a	6.36± 0.57	6.34± 0.47	6.40± 0.55
SOD (U/g)	103.00± 7.28	94.33± 5.74	100.67± 4.99	100.00± 3.61	106.00± 7.87
(MDA)+ 4- HNE (µmol/g)	1.55± 0.15	4.30 ± 0.28 a	2.34± o.10 b	1.57± 0.39 b	2.56± 0.03 ab

Mean ± SD of 6 animals in each group.
a Significant at P≤0.05 when compared with control group.
b Significant at P≤0.05 when compared with Al3+- intake group.

As shown in table (3): significant decreases of natural liver antioxidants GSH content; GSHpx and SOD activities in Al^{3+}- treated group. On the other hand, (MDA) + 4-HNE increased significantly in the same group when compared with control group. Rats treated with Al^{3+} and antioxidants showed improvements in the different tested parameters.Vitamin E had more antioxidant effect than M and NAC.

Table (3): some antioxidant parameters and {malondialdhyde(MDA) + 4- hydroxynonenal (4- HNE)}in the liver of Al^{3+}- treated rats and antioxidants (M, vit.E or NAC).

Parameters / Groups	control	Al^{3+}	Al^{3+}+ M	Al^{3+}+vit E	Al^{3+}+NAC
GSH (µmol/g)	21.94±1.90	9.39±13.9a	16.64±1.49b	37.49±2.08ab	15.95±2.12b
GSHpx (U/g)	581.983± 54.822	344.310± 51.371a	524.497± 72.770b	594.278± 53.272b	566.818± 65.268b
SOD (U/g)	46.143± 3.517	28.627± 5.065a	35.170± 4.771ab	46.188± 4.964b	40.063± 4.571b
(MDA)+ 4- HNE (µmol/g)	2.29± .22	4.24± 0.32 a	2.32± 0.16 b	1.29± 0.18 ab	2.49±0.07 b

Mean ± SD of 6 animals in each group.
a Significant at P≤0.05 when compared with control group.
b Significant at P≤0.05 when compared with Al3+- intake group.

Kidney GSH content and SOD activity decreased significantly while (MDA)+ 4- HNE level increased significantly (table :4)

Table (4): some antioxidant parameters and {malondialdhyde(MDA) + 4- hydroxynonenal (4- HNE)} in the kidney of Al^{3+}- treated rats and antioxidants (M, vit.E or ANAC).

Parameters / Groups	control	Al^{3+}	Al^{3+}+ M	Al^{3+}+ vit E	Al^{3+}+NAC
GSH (µmol/g)	23.66± 2.51	9.08± 0.96 a	15.66± 1.45 ab	29.89± 2.73 b	19.52± 2.53 b
SOD (U/g)	61.33± 3.65	30.68± 3.10 a	67.67± 1.96 b	72.00± 8.00 b	45.00± 3.17 ab
(MDA)+ 4- HNE (µmol/g)	1.57± 0.12	3.33± 0.09 a	2.79± 0.21 ab	1.25± 0.03 b	2.88± 0.17 ab

Mean ± SD of 6 animals in each group.
a Significant at P≤0.05 when compared with control group.
b Significant at P≤0.05 when compared with Al3+- intake group.

In the present study, testes GSH slightly decreased while SOD significantly decreased with a concomitant increase in the lipid end products in Al^{3+}- intake group. The use of external antioxidant vit, E , M and NAC combined with Al^{3+} enhanced these changes to nearly that of control group(table: 5).

Table (5): some antioxidant parameters and {malondialdhyde(MDA) + 4- hydroxynonenal (4- HNE)}in the testes of Al^{3+}- treated rats and Al^{3+} plus antioxidants (M, vit.E or ANAC).

Parameters / Groups	control	Al^{3+}	Al^{3+}+ M	Al^{3+}+ vit E	Al^{3+}+NAC
GSH (µmol/g)	27.38± 1.09	25.33± 0.38	27.27± 1.45	26.48± 0.74	26.41± 0.97
SOD (U/g)	28.00± 2.70	17.17± 1.40 a	26.33± 3.77b	31.33± 3.24 b	21.83± 1.42ab
(MDA)+ 4- HNE (µmol/g)	0.81± 0.04	1.92± 0.44 a	1.08 ± 0.06 b	0.69± 0.02 b	0.99± 0.09 b

Mean ± SD of 6 animals in each group.

a Significant at P≤0.05 when compared with control group.

b Significant at P≤0.05 when compared with Al3+- intake group.

Discussion

Aluminium is present in several manufactured foods and medicines and is also used in water purification (WHO, 2009). Aluminum is generally poorly absorbed by the gastrointestinal tract, much less than 1 percent in humans (Arnich *et al*., 2004 and Stella *et al*., 2005).

Aluminum has not been shown to have a definite biological function. Therefore, the present experiment was undertaken to determine the effectiveness of some antioxidants (M, vit. E or NAC) in modulating the aluminium chloride ($AlCl_3$) induced brain, liver, kidney and testes toxicity of rats.

Our results, recorded higher serum glucose and cholesterol levels in Al^{3+}- intake group when compared with control group. The increased level of serum cholesterol may be due to the increased lipid peroxidation and membrane fluidity which previously recorded by Silva *et al*. (2002). Many authors recorded similar results (El-Demerdash,2004 and Fyiad, 2007) who reported high levels of glucose and cholesterol in rats exposed to Al^{3+}. The antioxidants (M,vit.E or NAC) corrected the bad effects of Al^{3+} on serum glucose, cholesterol and triglycerides. These changes were returned to approximately normal levels by vit. E, melatonin, or NAC treatments which can be attributed to their antioxidant activity. Franzini *et al.*(2008) recorded lowered glucose levels by various antioxidants and attributed this results to their antioxidant actions.

In this study, highly significant increases of ASAT and ALAT were recorded. This agree with many authors who used Al^{3+} – oral administration (El-Demerdash, 2004), interperitonial or in drinking water (Nedzvetsky *et al.*, 2006). This elevations may be due to damage of cell membranes and release of its enzymes to the blood, since, Rajash and Latha, 2004) stated that elevation activities of these enzymes are indicative of cellular leakage and loss of the functional integrity of liver cell membrane. Also, Silva *et al*. (2002) and Stevanovic *et al*. (2008) suggested that the mechanism of Al^{3+} pro-oxidant action may be produced through its interaction with the membranes, subtle changes in the rearrangement of lipids which could attack and facilitate the propagation of lipid peroxidation leads to loss of membrane integrity, decrease its fluidity, disrupt the functioning membrane bound enzymes receptors and ion channels, which leads finally to cell death.

Oral administration of aluminium resulted in a significant increase in serum Al^{3+} of all Al^{3+}- exposed rats but not in tissues (brain, liver, kidney and testes) Aluminium contents of the studied tissues could not be detected in this study may be due to the different rout of administration and the small dose available for every rat or may be the detected limit of AAS system was high. Also, aluminium serum concentration in our study is only 8.443µg Al^{3+} / liter, Since antioxidants (vit E, M and NAC) have no effects on serum Al^{3+}- contents when combined with Al- exposure, it means that these antioxidants have no effect on aluminium execration.

Antioxidants are generally categorized to non-enzymatic and enzymatic. Non-enzymatic antioxidants include dietary compounds (vitamins C and E), minerals (selenium and zinc), glutathione, uric acid and

ubiquinol. Superoxide Dismutase (SOD), Catalase (CAT) and Glutathione Peroxidase (GPx) are the main enzymatic antioxidants (Abdollahi *et al*., 2004 and Rezaie *et al*., 2007).

In tissues such as liver and brain, GSH is oxidized to GSSG in the presence of ROS resulting in a shift of GSH. In the present study aluminium worthy acts as a pro-oxidant. Similarly, Esparza *et al*. (2003) suggested that aluminium might facilitate membrane peroxidation by increasing their susceptibility to free radicals induced damage.

Lipid peroxidation is one of the main manifestations of oxidative damage and has been found to play an important role in the toxicity and carcinogenicity of many xenobiotics (Anane and Creppy, 2001). Al^{3+} has been reported to induce lipid peroxidation, and to alter physiological and biochemical characteristics of biological systems. Experimental animal models and cell culture studies reveal that aluminium affects the expression of superoxide dismutase (SOD), catalase (CAT), glutathione peroxidase and glutathione (GSH) possibly leading to membrane fragility as a consequence (Abubakar *et al.*, 2003).

Elevation of lipid peroxidation in brain, liver, kidney, and testes as evidenced by the increased production of malondialdhyde (MDA) + 4- hydroxynonenal (4- HDNE) in the present study (tables:2,3, 4and 5), suggests participation of free – radical induced oxidative cell injury in mediating the toxicity of Al^{3+} as previously recorded by Anane and Creppy (2001) and Dua and Gill (2001). The aluminium-induced group had an increase in malondialdhyde (MDA) + 4- hydroxynonenal (4- HDNE) associated with a significant reduction ($P \leq 0.05$) in liver reduced glutathione levels and also a reduction of hepatic GSHpx and SOD activities. Furthermore, neurons appear to be particularly vulnerable to free radicals as the important natural antioxidant glutathione content is low, they have higher membrane content of polyunsaturated fatty acids and brain requires substantial quantities of oxygen for metabolism (Gupta *et al*., 2004).These changes were significantly attenuated in the Al^{3+} -exposed rats combined with antioxidants (M, vit. E or NAC). Since M, vit. E and NAC play important roles as antioxidants and are consequently expected to protect tissues from damage caused by reactive oxygen metabolites (El- Demerdash,2004, Prakash and Kumar (2009) and Esposito and Cuzzocrea (2010).Aydin *et al*. (2002) suggested that NAC decreased lipid peroxidation by direct scavenging of free radicals or by increasing GSH levels. In addition, Pocernich *et al*. (2001) and Kamboj *et al.* (2008) have also shown that NAC has an inhibitory effect on brain lipid peroxidation and has a protective role in membrane stabilization as a free radical scavenger.

Mohammadirad and Abdollahi (2011) reported that coadministration of α-tocopherol (Vitamin E) at 500 µg/g diet significantly preserved the GSH content of the brain and decreased the rate of lipid peroxidation. Brain had elevated lipid peroxidation end products {malondialdhyde (MDA) + 4- hydroxynonenal (4- HNE)} and reduction in GSH but not SOD activity. However, the lack of significant changes in brain cortex SOD activity (table, 2) after aluminium exposure is supported by the work of Abubakar *et al*. (2004b). These results may be due to the brain potent defenses against superoxide including dietary free-radical scavengers (ascorbate, α-tocopherol), the endogenous tripeptide glutathione, and enzymatic antioxidants (David *et al*. 2004).

Testicular oxidative stress appears to be a common feature in much of what underlies male infertility, which suggests that there may be benefits to develop better antioxidant therapies for relevant cases of hypospermatogenesis (Yousef and Salama, 2009 and Khattab *et al*., 2010). Yousef *et al.* (2005) found that aluminium enhanced lipid peroxidation in plasma, testes and liver.

In conclusion, the data presented in this paper using experimental animals, demonstrated that the toxic effects of Al^{3+} such as neuro -, hepato-, nephron- and testicular toxicity, as a result of oxygen free radical generation, can be alleviated by administration of antioxidants M, vit. E and NAC.

References

1. **Abdollahi M, Ranjbar A, Shadnia S, Nikfar S and. Rezaie A (2004):**Pesticides and oxidative stress: A review. Med Sci Monit, 10: RA141-RA147.
2. **Abubakar MGA, Taylor and Ferns G.A. (2003):** The effects of aluminium and selenium supplementation on brain and liver antioxidant status in the rat. Afri J Biotech; 3 (1): 88-93.
3. **Abubakar MGA, Taylor and Ferns G.A. (2004b):** Regional accumulation of aluminium

in the rat brain is affected by dietary vitamin E. J. Trace Elem Med Biol; 18: 53-59.

4. **Agency for Toxic Substances and Disease Registry (ATSDR), (1990):** Toxicological profile for aluminum. US Department of Health and Human Services. Public Health Service.
5. **Ammerman JP, Vhampman ML, Bouwman GW, Pontenot JP, Blagley CP and Moxan AI (1980):** Glutathione peroxidase. J. Animal Science; 51:1381.
6. **Anan, R and EE Creppy, (2001):** Lipid peroxidation as pathway of aluminium cytotoxicity in human skin fibroblast cultures: prevention bysuperoxide dismutase+catalase and vitamins E and C. Hum Exp Toxicol; 20: 477-481.
7. **Arnich N, Cunat L, Lanhers MC and Burnel D (2004):** Comparative in situ study of the intestinal absorption of aluminium , manganese, nickel and lead in rats. Biol. Trace Elem Res ; 99: 157- 171.
8. **Akyol A, Boyvat, A, and Kundakci N (2004):** Contact sensitivity to aluminium. Dermatology; 43:942-943.
9. **Aydin S, Ozaras R, Uzun H, Belce A, Uslu E, Tahan V, Altug T, (2002):** N-acetylcysteine reduced the effect of ethanol on antioxidant system in rat plasma and brain tissue. Tohoku J Exp Med ;198: 71–77.
10. **Becaria AA, Campbell and SC Bondy, (2002)**: Aluminum as a toxicant. Toxicol Ind Health; 18: 309-320.
11. **Bucolo G and David H (1973):** Quantitative determination of serum triglycerides by the use of enzymes. Clin Chem.; 19(5): 476-482. **Christen Y (2000):** Oxidative stress and Alzheimer disease. Am J Clin. Nutr; 71(2): 621s-629s.
12. **David S W, Sheng H and Batinic´-Haberle I (2004):**Review : Oxidants, antioxidants and the ischemic brain . J Exper Biol; 207:3221-3231.
13. **Dua R and KD Gill, (2001):** Aluminum phosphide exposure: implications on rat brain lipid peroxidation and antioxidant defence system. Pharmacol Toxicol; 89: 315-319.41.
14. **El- Demerdash FM (2004):** Antioxidant effect of vitamin E and selenium on lipid peroxidation , enzyme activities and biochemical parameters in rats exposed to aluminium. J. of Trace Elements in Med. And Biol; 18: 113 – 121.
15. **EPA (Environmental Protection Agency) (1997):** National ambient air quality standards for particulate matter; final rule. Fed Reg, 62:38651.
16. **Esparza J, Gómez M, Romeu M, Mulero M, Sánchez D, Mallol, J and Domingo J, (2003):** Aluminum-induced pro-oxidant effects in rats: protective role of exogenous melatonin. Journal of Pineal Research; 35 (1) : 32–39.
17. **Esposito E and Cuzzocrea S (2010):** Anti-inflammatory Activity of Melatonin in Central Nervous System . Curr Neuropharmacol; 8(3): 228–242.
18. **Esterbaur H, Schaur RJ And Zollner H, (1991):** Chemistry and biochemistry of 4-hydroxynonenal,and related aldhydes. Free radical. Bio Med; 4: 81-128.
19. **Farina** M, **Rotta** LN, **Soares** FAA , **Jardim** F ,**Souza DO and Rocha JBT (2005):** Hematological changes in rats chronically exposed to oral aluminum. Toxicology; 209(1): 29-37
20. **Franzini L , Ardigo D and Zavaroni I (2008):** Dietry antioxidants and glucose metabolism. Curr opin Clin Nutr Metab Care , 11: 471- 476.
21. **Fyiad A A (2007):** Aluminium Toxicity and Oxidative Damage Reduction By Melatonin in Rats J. Appl Sci Res; 3(10): 1210-1217.
22. **Goyer R, (2004):** The human health effects of metals. Issue paper submitted to ; U.S. Environmetal Protection Agency. Risk Assessment forum 1200 Pennsylvania Avenue, NW Washington, DC 20460, Contract # 68-C-02- 060.
23. **Gupta, YK, Sharma, M and Chaudhary, G (2004):** Oxidative stress in Alzheimer's Disease and central Eschemia: Implication for antioxidant treatment (book review) , Pharmacological perspectives of toxic chemicals and their antioxidants; (24): 431–442.
24. **Jansen S.; Broadley MR; Robbrecht E and Smits E(2002):** Aluminium hyperaccumulation in angiosperms : A review of its phylogenetic significance . The Botanical review; 68(2): 235-269
25. **Kamboj A, Kiran R, Sandhir R(2006a)**: Carbofuran-induced neurochemical and neurobehavioral alterations in rats: attenuation by N-acetylcysteine. Exp Brain Res; 170: 567–575.
26. **Kamboj A, Kiran R, Sandhir R(2006b)**: N-acetylcysteine ameliorates carbofuran-induced neurochemical alterations in lipid composition and activity of membrane bound enzymes. Mol Cell Biochem; 286:107-114.
27. **Kamboj, SS, Chopra, K, Sandhir, R (2008):** Neuroprotective effect of N-acetylcysteine in the development of diabetic encephalopathy in streptozotocin-induced diabetes. Metab Brain Dis; 23:427–443.
28. **Khattab HAH, Abdallah ZA and Kamel GM (2010):** Grape seed extract alleviate reproductive toxicity caused by aluminium chloride in male rats. J. Amer Scien; 6(12): 1200-1209.
29. **Krewski D, Yokel RA, Nieboer E, Borchelt, D, Cohen, J, Harry, J, Kacew S, Lindsay J,**

Amal, MM and Rond, V (2006): Human health risk assessment for aluminium , aluminium oxide and aluminium hydroxide. Dep. Epidemiology and community medicine , faculty of Medicine, University of Ottawa, Ottawa,Ontario, Canada. Email: cphra@uttawa.cs

30. **Lemire J, Mailloux R, Darwich R, Auger C and Appanna VD (2011):** The disruption of L-carnitine metabolism by aluminum toxicity and oxidative stress promotes dyslipidemia in human astrocytic and hepatic cells.Toxicol Lett; 203(3): 219-226.
31. **Lorenzoni AG and Ruiz-Feria A (2006):** Effects of Vitamin E and l-Arginine on Cardiopulmonary Function and Ascites Parameters in Broiler Chickens Reared Under Subnormal Temperatures. Poult Sci; 85 (12): 2241-2250.
32. **Mailloux RJ, Lemire J, Appanna VD (2011):** Hepatic response to aluminum toxicity: dyslipidemia and liver diseases. Exp Cell Res; 317(16):2231-8.
33. **Mohammadirad,A. and Abdollahi, M.(2011):** A Systematic Review on Oxidant/Antioxidant Imbalance in Aluminium Toxicity. International Journal of Pharmacology; 7: 12-21.
34. **Nishikimi M, Rao, NA and Yagl, K (1972):** The occurance of superoxide anion in the reaction of reduced phenazine methosulpharee and molecular oxygen. Biochem Biophys Res; 46: 844-853.
35. **Nedzvetsky, VS, Tuzcu, M, Yasar A, Tikhomirov AA and Baydas, G (2006):** Effects of vitamin E against aluminum neurotoxicity in rats. Biokhimiya; 71(3): 305–311.
36. **Paulis G; Ascenzo RD ; Nupieri P ; De Giorgio G ; Orsolini G ;Brancato T and Alvaro R (2011):**Effectiveness of antioxidants (propolis, blueberry, vitamin E) associated with verapamil in the medical management of Peyronie's disease:a study of 151 cases. Inter J Andrology; 6263:1-7.
37. **Prakash A and Kumar A (2009):** Effect of N-acetyl cysteine against aluminium-induced cognitive dysfunction and oxidative damage in rats.Basic Clin Pharmacol Toxicol; 105(2): 98-104.
38. **Pournourmohammadi S, Khazaeli, P, Eslamizad S, Tajvar A, Mohammadirad A and Abdollahi, M (2008):** Study on the oxidative stress status among cement plant workers. Hum. Exp. Toxicol; 27: 463-46923.
39. **Prins, HK and Loose, JA (1969):** Glutathione, In: " Biochemical methods in red cell genetics" Chapter 40.(edited by Yunis, J.J.), Academic press, N.Y. London pp. 126-129.
40. **Pocernich CB, Cardin AL, Racine CL, Lauderback CM, Butterfield DA(2001):** Glutathione elevation and its protective role in acrolein-induced protein damage in synaptosomal membranes: relevance to brain lipid peroxidation in neurodegenerative disease. Neurochem Int; 39: 141–149.
41. **Polizzi S, Pira E, Ferrara M, Bugiani M, Papaleo A, Albera R and Palmi S (2002):** neurotoxic effects of aluminium among foundry workers and alzheimer's disease. neurotoxicity; 23(6):n761-774.
42. **Quiles JL, Huertas JR, Battino M, Mataix J, Ramirez-Tortosa MC, (2002):** Antioxidant nutrients and adriamycin toxicity. Toxicology; 180,79-95.
43. **Quiles JL, Ochoa JJ, Huertas JR, Lopez-Frias M, Mataix, J (2006):** Olive oil and mitochondrial oxidative stress: studies on adriamycin toxicity, physical exercise and ageing. In: Quiles, J.L., Ramirez-Tortosa, M.C., Yaqoob, P. (Eds.), Olive Oil and Health. CABI Publishing, Oxford; :119–151.
44. **Rajash M and Latha M (200):** Preliminary evaluation of the antihepatotoxic effect of kailari, a polyherbal formulation. J.Ethnopharm; 91: 99-104.
45. **Ranjbar A K, Khani-Jazani A, Sedighi F, Jalali-Mashayekhi M, Ghazi-Khansari and M Abdollahi (2008):** Alteration of body total antioxidant capacity and thiol molecules in human chronic exposure to aluminum. Toxicol. Environ Chem; 90: 707-713.
46. **Reiter, RJ, Tan, DX, Qi W, Monchester LC, Karbownik, M and Calvi JR (2000):** Pharmacology and physiology of melatonin in the reduction of oxidative stress In vivo. Biol Signals Recept; 9: 160-71.
47. **Saiyed SM, Yokel RA. (2005**): Aluminum content of some foods and food products in the USA, with aluminum food additives. Food Addit Contam; 22(3):234-244.
48. **Schumann G; Bonora R; Ceriotti F; Férard G; Ferrero CA; Franck PFH; Gella FJ; Hoelzel W; Jørgensen PJ; Kanno T; Kessner A; Klauke R; Kristiansen N; Lessinger JM; Linsinger TP; Misaki H; Panteghini M; Pauwels J; Schiele F; Schimmel HG(2002):** IFCC primary reference procedures for the measurement of catalytic activity concentrations of enzymes at 37 °C: Part 4. Reference procedure for the measurement of catalytic concentration of alanineaminotransferase. Clin. Chem Lab Med; 40: 718-724
49. **Schumann G; Bonora R; Ceriotti F; Férard G; Ferrero CA; Franck PFH; Gella FJ; Hoelzel W; Jørgensen PJ; Kanno T; Kessner A; Klauke R; Kristiansen N; Lessinger JM; Linsinger TP; Misaki H; Panteghini M;**

Pauwels J; Schiele F; Schimmel HG; Weidemann G; Siekmann L(2002): IFCC primary reference procedures for the measurement of catalytic activity concentrations of enzymes at 37 °C: Part 5. Reference procedure for the measurement of catalytic concentration of aspartate-aminotransferase. Clin. Chem Lab Med; 40: 725-733.

50. **Silva SV, Cordeiro JM, Matos JM, Oliveira CR and Gonçalves PP(2002):** Aluminum accumulation and membrane fluidity alteration in synaptosomes isolated from rat brain cortex following aluminum ingestion: effect of cholesterol. Neuroscience Research; 44(2): 181-193.
51. **Soni MG, White SM and Flamm WG (2001)**. Safety evaluation of dietary aluminum. Regul Toxicol Pharmacol; 33(1):66-79.
52. **Sood PK, Nahar U and Nehru B (2011):** Curcumin attenuates aluminum-induced oxidative stress and mitochondrial dysfunction in rat brain Neurotox Res; 20(4):351-361.
53. **Stevanovic ID , Jovanovic MD , Jelenkovic A ,Colic M ,Stojanovic I and Ninkovic M (2008):**The effect of 7-nitroindazole on aluminium toxicity in the rat brain. Bulgarian J Veter Med; 11 (1)37- 47.
54. **Stella M, Ne'stor M, Marcela, G, Maria del Carmen C and Maria Monica, E (2005):** Alterations of the renal function and oxidative stress in renal tissue from rats chronically treated with aluminium during the initial phase of hepatic regeneration . J Inorganic Biochem; 99: 1858-1864.
55. **Tan DX, Manchester LC, Plummer BF, Limson J, Weintraub ST and Qi, W (2000):** Melatonin directly scavenges hydrogen peroxide: a potentially new metabolic pathway of melatonin biotransformation, Free Rad Biol Med; 29: 1177-1185.
56. . **Tietz N W (1995**): Clinical guide to laboratory tests. 3rd ed., W.B.Saunders Co., Philadelphia, 518-519.
57. **Vosters O, Neve J (2002):** Inhibitory effects of thiol-containing drugs on erythrocyte oxidative damages investigated with an improved assay system. Talanta; 57:595–600.
58. **World Health Organization, WHO (2009):** Chemical Hazards in Drinking Water-Aluminium(Online) last accessed 02.09.09 at http://www. who.int/water_sanitation_health/dwq/chemicals /aluminiumsum.pdf.
59. **Yokel, RA (2000):** The toxicology of aluminum in the brain: a review. Neurotoxicology; 21: 813-828.
60. **Yokel RA and McNamara PJ (2001):** Aluminum toxicokinetics: An updated mini review. Pharmacol Toxicol; 88: 159-167.3
61. **Yousef, M.I., El-Morsy, A.M.and Hassan, M.S. (2005):** Aluminum-induced deterioration in reproductive performance and seminal plasma biochemistry of male rabbits: Protective role of ascorbic acid. Toxicology; 215: 97-107.
62. **Yousef MI, Salama, AF (2009):** Propolis protection from reproductive toxicity caused by aluminium chloride in male rats. Food Chem Toxicol; 47(6):1168-75.

تحسين الشدة التأكسدية الناتجة من التسمم بالألومنيوم في ذكور الجرذان باستخدام بعض مضادات الأكسدة

أحكام محمود الجندي

كلية العلوم (بنات)جامعة الأزهر –القاهرة – مصر.

ملخص

يستخدم الألومنيوم بكثرة في أواني الطهي و التغليف وتنقية المياه وبعض العقاقير، كما قد يدخل الجسم عن طريق الجهاز التنفسي والجلد ؛ وقد تتولد شوارد حرة نتيجة للتسمم به. ولذلك تهدف هذه الدراسة لحماية الأنسجة (المخ والكبد والكلى والخصية) من التسمم بالألومنيوم باستخدام بعض مضادات الأكسدة (الميلاتونين وفيتامين هـ و ن- أسيتيل سيستيين).

تم إعطاء ذكور الجرذان البيضاء 53.5 مجم كلوريد الألومنيوم /لتر من ماء الشرب لمدة 30 يوما ؛ بينما عوملت باقى المجموعات بكلوريد الألومنويوم فى ماء الشرب بالإضافة إلى مضادات الأكسدة فيتامين هـ 100 مجم/كجم والميلاتونين 5مجم/كجم أو ن – أسيتيل السيستين 100مجم/كجم عن طريق أنبوب المعد ة وقورنت النتائج بالمجموعة الضابطة.

أرتفع مستوى الجلوكوز والكوليستيرول وإنزيمات الكبد ناقلات الأمين (ASAT & ALAT) ارتفاعاً ذو دلالة إحصائيةً فى مصل الدم؛ بينما انخفضت الدهون الثلاثية انخفاضا ذو دلالة إحصائية في المجموعة المسممة بالألومنيوم بالمقارنة بالمجموعة الضابطة . وتحسن هذا الانخفاض باستخدام مضادات الأكسدة: فيتامين هـ والميلاتونين و ن- أسيتيل سيستين .

وقد أظهر الجلوتاثيون المختزل في المخ والكبد والكلى والخصية نقصا ذو دلالة إحصائية في ذكور الجرذان المسممة بالألومنيوم عند مقارنتها بالمجموعة الضابطة ، وصاحبه نقص في الإنزيمات المضادة للأكسدة (جلوتاثيون بيروكسيديزفي الكبد وفوق أكسيد الديسميوتيز في الكبد والكلى فقط) ولم يتغير في نسيج المخ؛ وارتفاعا ذو دلالة إحصائية في أكاسيد الدهون الفوقية في جميع الأنسجة المختبرة. لعبت مضادات الأكسدة المستخدمة دورا هاما في تعديل هذه التغيرات وتحسينها إلى مستويات تقترب مما هو مسجل للمجموعة الضابطة .و كانت أفضل النتائج المسجلة لفيتامين هـ ثم الميلاتونين ويليه ن- أستيل السيستين.

ويستخلص من هذه الدراسة أهمية مضادات الأكسدة الطبيعية المتوفرة في الغذاء كفيتامين هـ , والعلاجية كالميلاتونين و ن- أستيل سيستيين لمنع حدوث مضاعفات التسمم بالألومنيوم الذي يستخدم بكثرة كأواني للطهي وعبوات غذائية و في بعض العقاقير

.