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# Effect of chronic aluminum chloride toxicity on sperm and reproductive markers in albino mice

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## Abstract

There is a little indication that aluminum is acutely toxic for the general population but prolonged exposure may induce systemic toxicity affecting the gastrointestinal tract, brain or skeleton. Therefore, the present study was conducted to investigate the effect of chronic aluminum chloride toxicity on sperm and some reproductive markers in Albino mice. A total of 24 mice each were injected an IP sub-chronic dose of 55.45mg/kg IP, or saline (control) daily for 60 consecutive day. After the 7th, 15th and 30ty 60ty day of dosing, mice were sacrificed for sperm counts and quality and microscopy of testis, prostrate and epididymis. The study showed that the effect increased with increasing ALCl3 dose and duration of exposure. The study also showed significant ($P<0.05$) increasing in the rates of birth defects in sperm with a significant decrease ($P <0.05$) in the preparation of her natural and special periods (30,60) for the chronic toxic Exposure. As explained microscopic examination of scarified mice ranged from pathological lesions which was getting over exposed began toxic degeneration of cellular necrosis in the cells of members with hemorrhage and congestion and the disappearance of the evolution of sperm, and infiltrates of inflammatory cells (lymphocytes and macrophages).

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## Introduction

Aluminum(Al) is found in about 8% of the Earth's crust in combination with oxygen, silicon, fluorine and other elements in soil, rocks, clay and gems (Casarett and Doll's, 2003) but biological function was not clearly known (Farina *et al.,* 2002). Aluminum is found in corn, yellow cheese, salts, herbs, spices and tea (Casarett and Doll's, 2003).

Al is used in cosmetics, antiperspirants, deodorants, antacids, buffered aspirin, antidiarrheals, vaccines and allergen injections. Also Al is used in the veterinary medicine, glues and disinfectants (Kachny *et al.,* 1997; Yousef, 2004). There is a little indication that aluminum is acutely toxic for the general population but prolonged exposure may induce systemic toxicity affecting the gastrointestinal tract, brain or skeleton (ATSDR, 1997).

Chronic AL toxicity occurs almost exclusively in persons undergoing dialysis for renal failure, who are likely to develop osteomalacia or aplastic bone disease, respiratory diseases (e.g., pulmonary fibrosis, occupational or potroom asthma, and chronic bronchitis), neurological effects, including impairment of cognitive function and motor dysfunction, Alzheimer's disease, amyotrophic lateral sclerosis (ALS) and Parkinson's disease, (Golub and Germann, 1997). $AlCl_3$ induced reproductive toxicity and adverse effect on the steroidogenesis (Yousef *et al.*, 2005) ,with alterations in the metabolism of testis, epididymis and vas deferens that led to poor sperm motility and reduction in fertility rate in mice treated with ALCL3(Yousef, 2009;Chinoy, 2001) ,Also oral demonstration of $ALCl_3$ in dose (200)mg/kg body weight for 60 days in chronic treatment lead to severe damage within seminiferous tubules and vascular degeneration on spermatogonic and Sertoli cells cytoplasm (Khattab, 2007). Adebayo *et al.,* (2012) found that oral administration of $AlCl_3$ to wistar rats of doses (475) mg/kg .bw (950) mg/kg.bw, (1425) mg /kg .bw and (1900) mg/ kg.bw noted that histological observations revealed that there were no significant histological changes effects. Agrawal *et al.,* (1996) noticed that histopathological examination revealed degenerative changes of the seminiferous tubules with focal areas of necrosed spermatogenic cells, marked degeneration and desquamation of the lining epithelial cells of epididymis as well as multiple calcified materials in prostate gland following 60 days of aluminium treatment. The studies of the effect of chronic aluminum chloride toxicity on sperm and some reproductive markers therefore this study was carried out to investigate the its effect in mice.

## Materials and methods

Male mice (n=48) aged 3 months and weighing 30–35g from the animal house of the Embryo Research and Infertility Treatment Institute, Al-Nahrain University, Iraq, were used. Mice were acclimatized in plastic cages 30*10*10 $cm^3$ for 2 weeks. Standard rodent diet (Commercial feed pellets) and drinking water were given. Housing conditions were maintained at 22± 4°C, the air of the room was changed continuously by using ventilating vacuum and light/dark cycle (14/10) hr/per/day.

The experiment was conducted in the animal house of the Pathology Department, College of Veterinary Medicine, Baghdad University, Iraq.

The dose of sub chronic of AlCl3 was estimated according to Mohammad, 2014, Groups of 24 mice each were injected a low dose of 55.45 mg/kg AlCl3 IP daily for 60 days. Control group (n=24) was injected an IP of 55.45 mg/kg of normal saline daily. After the 7th, $15^{th}$, 30ty, and 60ty day dosing, mice were scarified for sperm counts/quality and microscopy.

Sperm was obtained by cutting of the *cauda epididymis* with surgical blades and squeezed into a sterile clean watch glass as described by Bowman (2000). For progressive motility and concentration, sperm was mixed and diluted 10 times in 2.9 % sodium citrate dehydrate solution. A drop of suspension was smeared on a glass slide and stained by eosin blue stain to determine percentage of viable sperm and morphology examined under macroscopic immersed in oil Bowman (2000).

Mice were sacrificed at 7th, 15th, 30ty, and 60ty day (6 mice for control groups and groups given Alcl3) by chloroform inhalation, for microscopy of testis, epididymis and prostate gland. Macroscopic changes were recorded.

Tissues were fixed in 10% formaldehyde and processed by histokinette as described by Luna and Lea, (1968). Least significant difference (LSD) tests was used to compare differences between means with SAS, (2012).

**Results**

By day 7, sperm counts in mice given Aluminum chloride declined ($P < 0.05$) between by day 7 (3.12 ± 0.12), (2.25 ± 0.16) & control group respectively while the most significant decrease was recognized at day 15 (3.00±0.013), (2.25 ± 0.16) at day 30 (2.50±0.18), (0.62 ± 0.18) and at day 60 (2.75 ± 0.16), (0.37 ± 0.18) as in Table1.

**Table 1.** Sperm counts (1000000/ ml) in mice given 55.45 mg/kg aluminum chloride Daily for 60 days and saline controls.

| Period (Day) | The Group | | LSD value |
|---|---|---|---|
| | Control | Sub Chronic toxic | |
| 7 | 3.12 ± 0.12A a | 2.25 ± 0.16 B a | 0.81 * |
| 15 | 3.00 ± 0.13 A a | 2.25 ± 0.16 B b | 0.577 * |
| 30 | 2.50 ± 0.18 A a | 0.62 ± 0.18 B c | 0.779 * |
| 60 | 2.75 ± 0.16 A a | 0.37 ± 0.18 B d | 0.782 |
| LSD value | 0.636 NS | 0.811 * | --- |

Means with a different small letter in the same column significantly different ($P<0.05$), Means with a different capital letter in the same row significantly different ($P<0.05$).

At day 7 we found no differences in live sperm between treated and control mice. However by day 15 , day 30 and day 60, decreased ($P < 0.05$) in those given Aluminum Chloride as Shown in Table 2, below. We found abnormal sperm morphology in mice given aluminum chloride Throughout compared with control from day 7 of $ALCl_3$ in sub-chronic toxic effect Showed a significant increase ($P< 0.05$) at days 15, 30 mostly at day 60 compared to Control group (8.37 %), &sub-chronic toxic effect (21.87 %) Table 3.

**Table 2.** Live sperm (%) in mice given 55.45 mg/kg aluminum chloride or saline (Controls) daily for 60 days.

| Period (Day) | | | LSD value |
|---|---|---|---|
| | Control | Sub chronic toxic | |
| 7 | 85.00 A a n=6 | 80.87 A a | 6.85 NS |
| 15 | 84.75 A a n=6 | 77.70 B a | 5.91 * |
| 30 | 83.12 A a n=6 | 65.25 B b | 8.24 * |
| 60 | 83.75 A a n=6 | 50.00 B c | 8.96 * |
| LSD value | 0.636 NS | 0.811 * | --- |

Means with a different small letter in the same column significantly different ($P<0.05$). Means with a different capital letter in the same row significantly different ($P<0.05$).

At day 30 there were secondary abnormalities in the sperm different, amorphous head, hookless, banana, double-headed, coiled with microcephaly, bent at cephalocaudal junction, bent with projecting filaments, microcephaly with tail defect; and defective head with duplication of tail. Like but mostly we have bent tail & defect in the head of sperm as in Fig. 1 & 2.

**Table 3.** Abnormal sperm (%) in mice given 55.45 mg/kg aluminum chloride daily For 60 days and in saline controls.

| Period (Day) | The Group | | LSD value |
|---|---|---|---|
| | Control | Sub chronic toxic | |
| 7 | 8.87 A a | 8.08 A d | 1.142 NS |
| 15 | 8.87 B a | 14.25 A c | 2.087 * |
| 30 | 8.62 B a | 17.75 A b | 2.362 * |
| 60 | 8.37 B a | 21.87 A a | 3.819 * |
| LSD value | 1.075 NS | 2.37 * | --- |

Means with a different small letter in the same column significantly different (P<0.05) Means with a different capital letter in the same row significantly different (P<0.05).

We found macroscopic lesions in Sub chronic toxicity groups which characterized by enlargement of testis in mice as shown in Fig 3.

**Fig. 1.** Defect in the head of mice sperm sub chronic Low toxic dose (55.45)mg/kg.bw at 30 days (Eosin X100).

Microscopy showed no changes by day 7 but by day 15; in mice given $AlCl_3$ we observed, in testis fibrin deposition, the lesion of testicular tissue showed sever vacuolization of semineferous tubules with hyalinization together with interstial proliferation of lydic cells as well as slight fibrous thickness of tunica albugina ,while other section , Showed proliferation of lydic cell associated with slight interstial fibrosis**,** There was slight fibrosis in tunica albugina associated with lack of spermatocyte in the lumen of some semineferous tubules. the testicular lesion showed increase thicking of tunica albugina other section showed sloughing off of spermatogenic cell in the lumen of semineferous tubules (S.T) with sever necrosis together with interstitial fibrosis also the result showed focal mononuclear cells infiltration and blood vessels congestion with fibrosis seen in surrounding testicular tissue, Diffuse cellular aggregation in the paratistecular tissue with moderate fibrosis and no clear lesion in semineferous tubules while other section showed marked distension of semineferous tubules ( S.T) that lack & devoid of spermatid with slight fibrosis of tunica albugina .Also result showed giant cell formation in some semineferous tubules accompanied with diffuse necrosis (Figs4,5,6A)

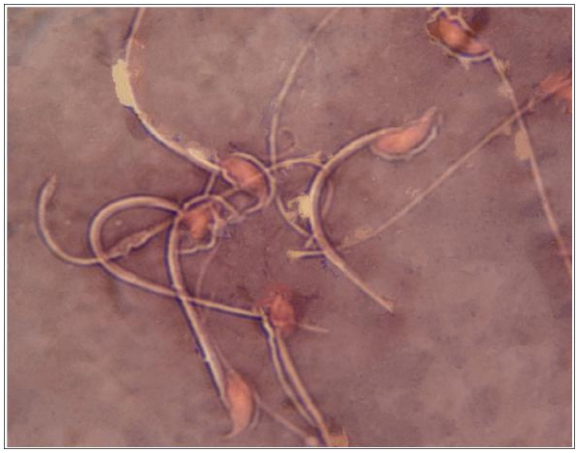

**Fig. 2.** Secondary abnormalities in sperm (bent) in mice revealed Alcl3 sub chronic low toxic dose Stain(55.45) mg /kg.b.w for 60 days (Eosin stain X100).

Epidiymis**:** The main feature of epididymal tissue at this period characterized by sever destruction of ductal epididymal epithelia together with mild interstitial fibrosis & mild mononuclear cells infiltration, higher magnification of above section showed interstial fibrosis with focal mononuclear cells infiltration in the epididymal tissue.

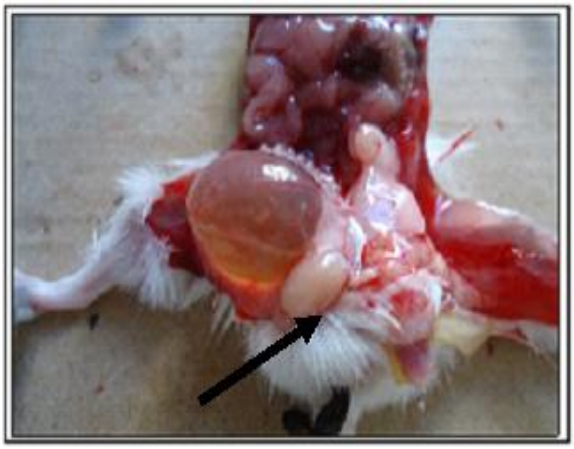

**Fig. 3.** Macroscopic appearance of testis mice low sub chronic Toxic effect dose (55.45)mg/kg.bw I/P at day 60 showing Enlargement of testis marking.

Epididymal tissue showed interstial fibrosis mixed with slight mononuclear cells infiltration together with spermatid loss in some tubules & spermatid cloumping in others, Sever destruction in the epithelial of epididymis (caput) associated with sloughing & slight cellular infiltrate together with moderate fibrosis as well as mononuclear cells infiltration in L.P mainly around blood vessels (Figs4,5,6B).

Prostate**:** There was fibromuscular proliferation in prostate stromal tissue with slight mononuclear cells aggregate together with sever epithelial hyperplasia (Figs 4, 5, 6C).

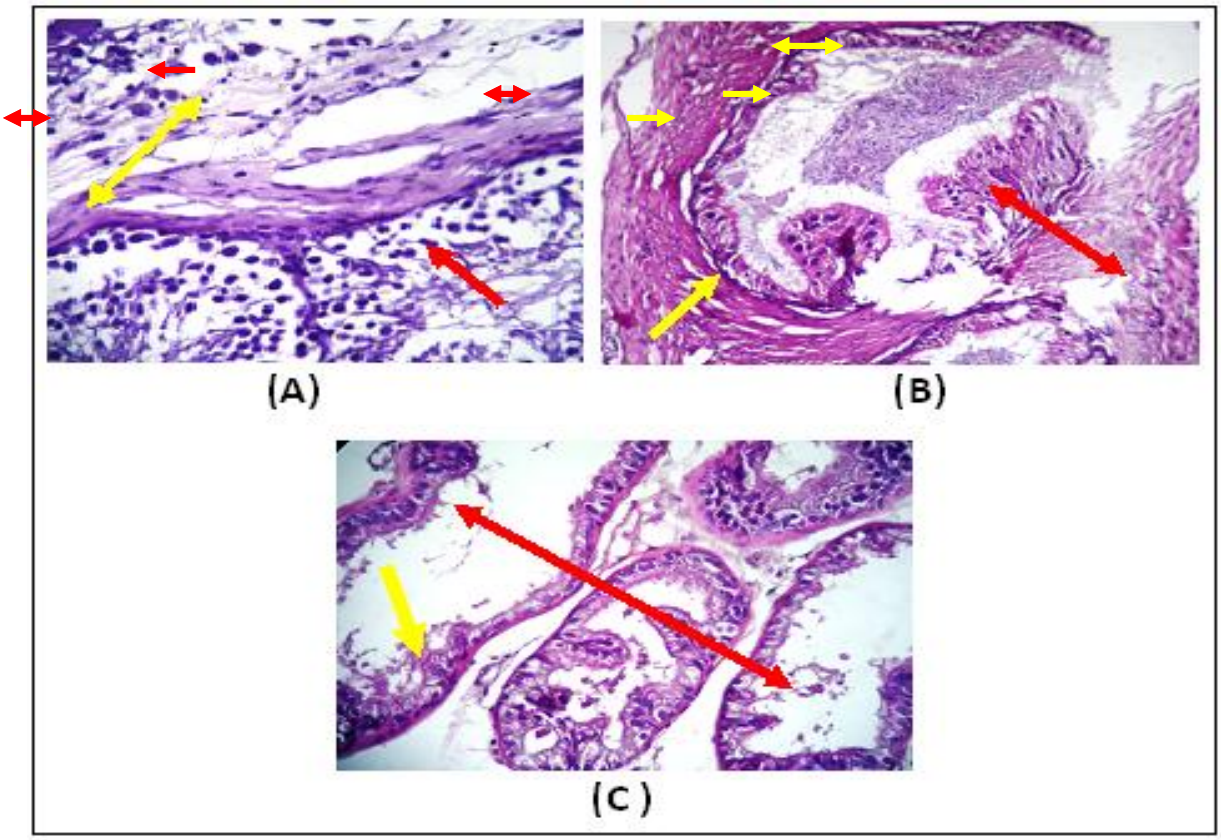


**Fig. 4.** (A, B, C): Histopathological section in (A) testis (B) epidiymis (C) prostate of low chronic toxic dose at 15 days shown (A) Fibrin deposition & MNCs cellular deposition (macrophage & plasma cell) in tunica albugina ↔ with disruption of spermatied in seminferous tubules ← (H&E stain X40),(B) Moderate to sever destruction in the epithelial lining with sloughing & slight infiltration with mononuclear cells infiltration in the interstial tissue ↔ as well as fibrosis → (H&E stain X40).(C) Cystic distension of prostate acini ↔ together with desquamated epithelial that exhibited in others →(H&E stain X40).

**Discussion**

According to toxicity rate the $LD_{50}$ of $AlCl_3$ which was given to mice through I/P injection considered moderately toxic since $LD_{50}$ we found $AlCl_3$ is (1109.17)mg/kg. bw in mice and considered as highly toxic compound. Our results are disagreed with other

researches, (WHO, 1997) were showed that the range of $ALCl_3$ $LD_{50}$ was (25-82) mg/kg.bw in mice while (Alleva *et al.,* 2005) showed that the $LD_{50}$ of $ALCl_3$ in white mice was (54)mg/kg .bw. The present in this experiment revealed that there was a statistical significant decreased in sperm count of $ALCl_3$ toxic effect groups when compared with the control and the displayed decreased was statistical significant at ($P<0.05$),this evidence was (Liboet *et al.,* 1995; Colomina *et al .,* 1998).low sperm count is the most frequent cause of male infertility (Buraimoh *et al.,* 2012a).Over exposure to environmental assaults (toxins chemicals and infections)can reduce sperms count(Ranjbar *et al.*,2009).

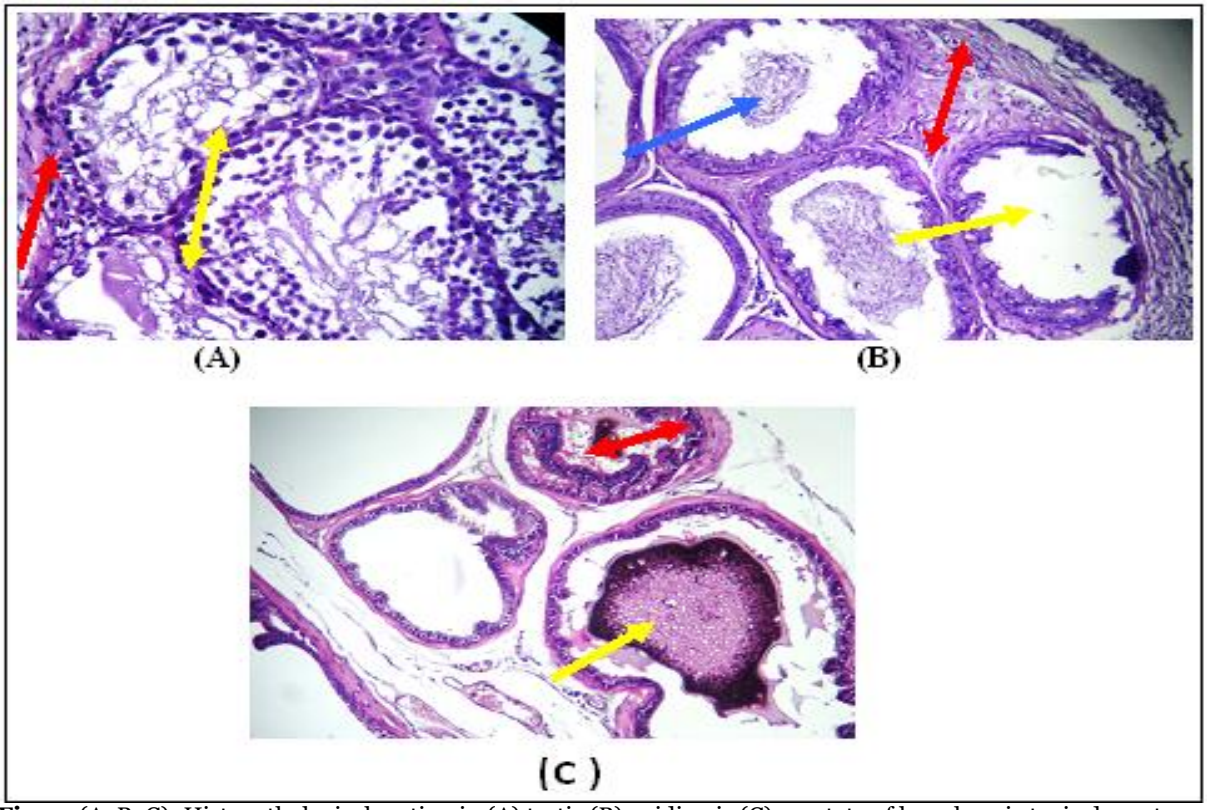


**Fig. 5.** (A, B, C): Histopathological section in (A) testis (B) epidiymis (C) prostate of low chronic toxic dose at **30** days shown: (A) Sever necrosis & hylainzation of seminferous tubules with interstial proliferation of lydig cell as well as slight fibrous thicking of tunica albugina (H&E stain X40), (B) Interstial fibrosis mixed with slight MNCs cell infiltration together with spermatied loss in some tubules & spermatied clumping in others (H & Estain X10) , (C) Great cystic distenstion of prostate acini with mineralization of acinar secretion & degenerative changes in some acini (H & E stain X10).

The administration of $AlCl_3$ caused a significant decrease in both sperm counts and live sperm percentage, with associated significant increase in the percentage of abnormal sperms as compared with the control groups, which those results obtained by other investigators (Liobet, 1995; Mayyas *et al.,* 2005; Yousef *et al.,* 2005). These changes may be attributed to impairment of sperm maturation and secretary functions of epididymal cells which might be due to oxidative stress or to insufficiency of androgens. Recent studies showed the AlCl3 caused a significant induction of oxidative stress as evidenced by increasing thiobarbituric acid reactive substances (TBARS) levels and inhibition of superoxide dismutase and catalasea ctivities (Yousef *et al.,* 2005; Sharma *et al.,* 2007)and decreased glutathione Furthermore, the defect in the head of sperm & secondary abnormalities in sperm and abnormal morphology, was similar to observation by Bray *et al.,* (2012) and Pizent *et al.,* (2012). El-Demerdash (2004) found there was significant decreased in the sulphydryl groups and phosphorylase in rats treated

with $AlCl_3$ so that a generation of reactive oxygen species (ROS) which may stimulates the glutathione peroxidase and inhibits catalase which result in oxidation of proteins in the terminal stages of sperm maturation (Maiorino, 2002), in addition to peroxidation of polyunsaturated fatty acids in the plasma membrane of sperm and this causes disturbances in sperm functions.(Gil-Guzman *et al.,* 2001; Ollero *et al.,* 2001; Orihuela *et al.,* 2005) showed that levels of reactive oxygen species production in semen were negatively correlated with the percentage of normal sperm forms as determined by ( Goel *et al.,*1997; Flten, 2001).

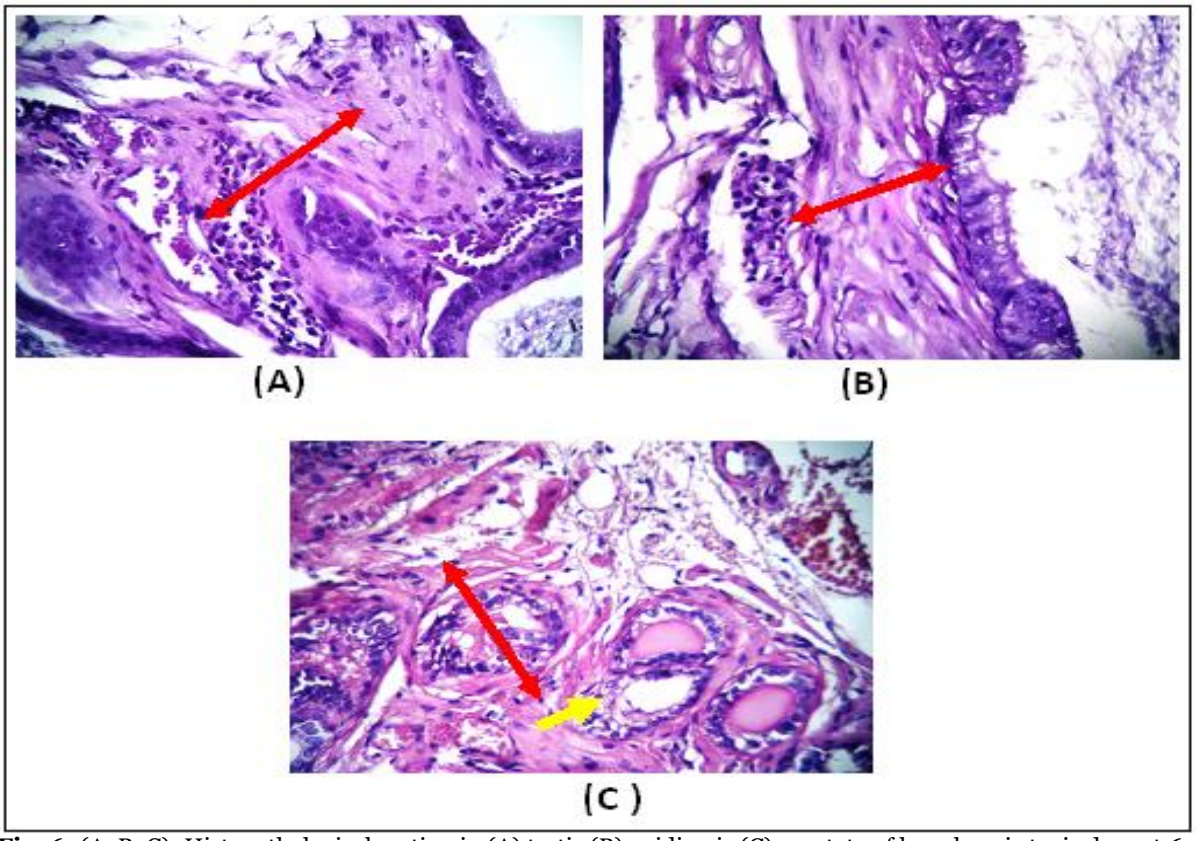


**Fig. 6.** (A, B, C): Histopathological section in (A) testis (B) epidiymis (C) prostate of low chronic toxic dose at **60** days shown: (A) Focal moderate mononuclear cells infiltration and blood vessels congestion with fibrosis in the surrounding testicular tissue↔ (H&E stain X40) ,(B) Sever fibrosis with interstail focal mononuclear cells infiltration in the epididymal tissue ↔ (H & E stain X40) (C) Fibroplasia of prostate interstial tissue with slight cellular aggregate → some acini exhibdated epithelial hyperplasia & other appearance normal ↔(H&E stain X40).

In general result of the present study indicates that there was a relationship between oxidative stresses induced by aluminum chloride toxicity (Acute low toxic effect& chronic high toxic effect) and increased the percentage of morphologically abnormal sperms and death in the total sperm count. The present gross pathological lesions showed that the animals exposed to toxic doses according to the dose exposure to $ALCl_3$ depends on the toxic effect dose and duration of injection& mainly seen at day 60 that macroscopic appearance of organ in chronic toxicity was characterized by severe congestion of uterus and oviduct and that reflect the severe vascular effects of $ALCl_3$ as similar disrupt blood brain barrier permentability (Jones and Hunt, 1983) Similar results have been reported by Summaedaey, (1989). The present results demonstrated several changed with in the interstitial leyding cell with slight proliferation mainly at (15), (30), (60) day with (55.45)mg/kg.bw was in consistence with (Issam *et*

*al.*, 2005) record that Ingestion of $ALCl_3$ has direct effect on testicular leyding cells in which testosterone is produced and $ALCL_3$ have effected the function of hypothalamus ,this may result alteration of gonadotropin reasing hormone (GnRH) levels from the anterior pituitary lobe which in turn will effect leyding & sertoli cells thus altering testosterone production.

The presented recorded gaint cells formation at (30), (60) days with (55.45) mg/kg.bw, result is agreement with with (Libet *et al.*,1995)who although found degenration in structure of spermatogenosis & formation of gaint cells . Our study showed several pathological lesion in prostate characterized mainly by marking acini epithelial hyperplasia mainly at (14) day with (221.83)mg/kg.bw post treatment similar with observed by AL ions replace iron and magnesium ions resulting in the reduction of Fe 2+ to ferretin. Free iron released from biological complexes by Al can catalyze hydrogen per oxide decomposition to hydroxyl radical. This high hydroxyl radical reactivity is able to initiate lipid peroxidation (Ward *et al.* 2001) there were no histopathological changes in prostate gland in reported agree or disagree on the prostate gland.

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**Reference**

**Adebayo AB, Samuel AO, Joseph OH, Sunday SA.** 2012. Aluminium chloride exposure had no effects on the epididymis of Wistar rats. American Medical Journal **3(2),** 210-219.

**Agrawal SK, Ayyash L, Gourley CS, Levy J, Faber K, Hughes CL.** 1996. Evaluation of the developmental neuroendocrine and reproductive toxicology of aluminium. Food Chemical Toxicology **34,** 49-53.

**Alleva E, Rankin J, Santucci D.** 1998. Neurobehavioral alteration in rodents following developmental exposure to aluminum. Toxicology and Industrial Health **14(1–2),** 209 – 221.

**ATSDR.** 1997. Toxicological Profile for Aluminum (Update). Draft for public comment. Agency for Toxic Substances and Disease Registry, Public Health Service, U.S. Department of Health and Human Services. Atlanta, GA.

**Bownan WC, Rand MJ.** 2000. Text Book of Pharmacology, 2nd ed., Bowman and William Cameron, Great Britain by University press, Cambridge

**Bray JJ, Cragg PA, Macknight ADC, Mills RG.** 2012. Reproduction, In: Huamn physiology, Bray, J.J., Cragg, P.A., Mack night, A.D.C. and Mills, R.G. (eds), 4th ed., Blackwell Science, USA, Pp, **274**.

**Buraimoh AA, Ojo SA, Hambolu JO, Adebisi SS.** 2012a. Brain aluminium uptake of adult wistar rats following the oral administration of aluminium chloride. European Journal of Scientific Research **72,** 576-582.

**Chinoy NJ, Trupti N, Patel A.** 2001. Effects of sodium fluoride and aluminium chloride on ovary and uterus of mice and their reversal by some antidotes. Fluoride **34(1),** 9-20.

**Colomina MT, Esparza JL, Corbella J, Domingo JL.** 1998. The effect of maternal restraint on developmental toxicity of aluminium in mice. Neuro toxicology and Teratology **20**, 651-656.

**El-demerdash FM.** 2004. Antioxidant effect of vitamin E and selenium on lipid peroxidation, enzyme activities and biochemical parameters in rats exposed to aluminum. Journal of Trace Elements and Medical Biology **18,** 113-121.

**Gil-Guzman E, Ollero M, Lopez MC, Sharma RK, Alvarez JG, Thomas AJ Jr, Agarwal A.**

2002. Differential production of reactive oxygen speciesby subsets of human spermatozoa at different stages of maturation. Human Reproduction **16,** 1922–1930.

**Farina M, Lara FS, Brandao R, Jacques R, Rocha JBT.** 2002. Effects of aluminum sulfate on erythropoiesis in rats. Toxicology Letter **132**, 131-139.

**Flaten R.** 2001. Absorption of aluminum from antiperspirants and vaccine adjuvants. In″ Aluminum and Alzheimer's Diseases : The Science that Describes the link. ″ (C. Exley, Ed), p 75–95. Elsevier. Amsterdam.

**Goel KA, Sastry KV.** 1997. A Text Book of Animal Physiology, Rakesh Kumar Rastogi, India p, **443**.

**Golub MS, Germann SL.** 1997. Aluminum effects on operant performance and food motivation of mice. Neurotoxicology and Teratology **20(4),** 421 – 427.

**Issam M, Ahmed E, Khamas WA.** 2005. Evaluation of reproductive and fertility toxic potential of ALCL3 on adult male mice. Journal of Animal and Veterinary Advances **4(2),** 224-233.

**Jones TC, Hunt RD.** 1983. Veterinary Pathology Leo & Febiger. Philadelphia, p **167**.

**Kachny W, Hegg A, Alfrey A.** 1997. Gastrointestinal absorption of aluminium from aluminium containing antacids. New England Journal of Medicine **296,** 1389-1390.

**Khattab IKF.** 2007. Histological and ultra structural studies on the testis of rat after treatment with aluminium chloride. Australian Journal of Basic Applied Science **1,** 63-72.

**Liobet JM, Colomina MT, Sirvent JJ, Domingo JL, Corbella J.** 1995. Reproductive toxicology of aluminium in male mice. Fundamental Applied Toxicology **25 (1),** 45-51 .

**Luna GL.** 1968. Manual of Histologic staining methods of the armed forces Institute of Pathology. 3rd edition. McGraw-Hill Book Company, New York, USA.

**Mayyas I, Elbetieha A, Khamas W, Khamas A.** 2005. Evaluation of reproductive and fertility toxic potentials of aluminium chloride on adult male mice, Journal of Animal and Veterinary Advances **4(2),** 224-233.

**Mohammad AM.** 2014. Toxicopathological Effect of Aluminum Chloride (ALCL3) in Male and Female Reproductive System of Albino Mice. M. Sc. thesis, College of Veterinary Medicine Baghdad University.

**Ollero M, Gil-Guzman E, Lopez MC, Sharma RK, Agarwal A, Larson K, Evenson D, Thomas AJJR, Alvarez JG.** 2001. Characterization of subset of human spermatozoa at different stages of maturation: implications in the diagnosis and treatment of male infertility. Human Reproduction, **16,** 1912–1921.

**Pizent A, Tariba B, Živković T.** 2012. Reproductive toxicity of metals in men, Archives of Industrial Hygiene and Toxicology **63,** 35-46

**Ranjbar AR, Khani-Jazani A, Sedighi F, Jalali-Mashayehi M, Ghazi-Khansari, Abdollahi M.** 2008. Alteration of body total antioxidant capacity and thiol molecules in human chronic exposure to aluminum. Toxicology and Environmental Chemistry **90,** 707-713.

**SAS.** 2012. Statistical Analysis System, User's Guide. Statistical. Version 9.1th ed. SAS. Inst. Inc. Cary. N.C. USA.

**Sharma P, Shah ZA, Kumar A, Islam F, Mishra KP.** 2007. Role of combined administration of Tiron and glutathione against aluminum-induced oxidative stress in rat brain. Journal of Trace Elements and Medical Biology **21**63–70.

**Summaedaey AS.** 1989. Experimental Study of Mercurical Toxicosis in Rats. Iraqi Journal of Veterinary Med **36(2),** 231– 243.

**Ward RJ, Zhang Y, Crichton RR.** 2001. Aluminum toxicity and iron homeostasis. Journal of Inorganic Biochemistry **87(1-2)**, 9-16.

**WHO.** 1997. Aluminium. Geneva, World Health Organization, International Programmer on Chemical Safety (Environmental Health Criteria **194.**

**Yousef MI.** 2004. Aluminum-induced changes in hemato-biochemical parameters, lipid peroxidation and enzyme activities of male rabbits: protective role of ascorbic acid. Toxicology **199,** 47-57.

**Yousef MI, Salama AF.** 2009. Propolis protection from reproductive toxicity caused by aluminium chloride in male rats. Food Chem. Toxicology **47,** 1168-1175.

**Yousef MI, El-Morsy AM, Hassan MS.** 2005. Aluminum-induced deterioration in reproductive performance and seminal plasma biochemistry of male rabbits: Protective role of ascorbic acid. Toxicology **215,** 97-107.
http://dx.doi.org/10. 1016/j.tox. 06. 025.