Histological Study of the Effects of Aluminium Chloride Exposure on the Testis of Wistar Rats

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Abstract

Aluminium compounds are used in pharmaceuticals and in water treatment processes.Testis is one of a pair of organs(pleural is Testes)and are site of sperm production.This study was aimed at evaluating the effects that aluminium chloride exposure could have on the testis of wistar rats. The wistar rats were divided into five groups:Group I was the control that received distil water while Groups II to V were given various concentrations of Aluminium chloride.After eight weeks of oral administration,the wistar rats were humanely sacrificed and the testes were removed and fixed in bouin's fluid.The testes were processed and stained in Haematoxylin and Eosin.The histological observations revealed seminiferous tubules that attained different shapes, vacuolar cytoplasm with loss of normal distribution of the epithelial linning in treated groups when compared with control group.We therefore conclude that aluminium chloride exposure could be detrimental to the integrity of the testes of wistar rats.

Key words: Histological study, Effects, Aluminium Chloride, Testis, Wistar Rats.

1. Introduction

Testis is one of the pair of male gonads that produces sperm and testosterone. The adult testes are suspended in the scrotum by the spermatic cords; in early fetal life they are contained in the abdominal cavity behind the peritoneum. Before birth, they normally descend into the scrotum.

The coverings of the testes are the skin and the dartos tunic of the scrotum, the external spermatic fascia, the cremasteric layer, the internal spermatic fascia, and the tunica vaginalis. Each testis is a laterally compressed oval body about 4 cm long and 2.5 cm wide that weighs about 12 g. It is positioned obliquely in the scrotum, with the cranial extremity directed ventrally and slightly laterally and the caudal end directed dorsally and slightly medially. The anterior border, lateral surfaces, and extremities of the organ are convex, free, smooth, and covered by the tunica vaginalis. The convoluted epididymis lying on the posterior border of the testis contains a tightly coiled tube that is about 20 feet long and connects with the vas deferens through which spermatozoa pass during ejaculation. Each testis consists of several hundred conical lobules containing the tiny coiled seminiferous tubules, each about 75 mm long, in which spermatozoa develop. In early life, the tubules are pale in color, but in old age, they become invested with yellow fatty matter. The tubules converge to form the rete testis, which is drained by the efferent ducts into the head of the epididymis.

The testes are supplied with blood by the two internal spermatic arteries that arise from the aorta, are served by the testicular veins that form the pampiniform plexuses constituting the greater part of the spermatic cords, and are innervated by the spermatic plexuses of nerves from the celiac plexuses of the autonomic nervous system. (Gray, 2000; Mosby's Medical Dictionary).

Almost all healthy male vertebrates have two testes. Plural testes, also called testicle, in animals, the organ that produces sperm (spermatogenesis), the male reproductive cell, and androgens, primarily testosterone, the male hormones. In humans, the testes occur as a pair of oval-shaped organs. They are contained within the scrotal sac, which is located directly behind the penis and in front of the anus. Both functions of the testicle are influenced by gonadotropic hormones produced by the anterior pituitary. Luteinizing hormone (LH) results in testosterone release. The presence of both testosterone and Follicle-Stimulating Hormone (FSH) is needed to support spermatogenesis (Michael and Wojciech, 2006; Romer and Parsons, 1977; Skinner *et al*., 1989). There are two phases in which the testes grow substantially; namely in embryonic and pubertal age (Scott, 2000). The testes grow in response to the start of spermatogenesis. The testes contain germ cells that differentiate into mature spermatozoa, supporting cells called Sertoli cells, and testosterone-producing cells called Leydig (interstitial) cells. The germ cells migrate to the fetal testes from the embryonic yolk sac. The Sertoli cells, which are interspersed between the germinal epithelial cells within the seminiferous tubules, are analogous to the granulosa cells in the ovary and the Leydig cells, which are located beneath the tunica albuginea, in the septal walls, and between the tubules, are analogous to the hormone-secreting interstitial cells of the ovary (Gray, 2000).

Aluminum is a trivalent cation found in its ionic form in most kinds of animal and plant tissues and in natural waters everywhere (Jiang, et al., 2008). It is the third most prevalent element and the most abundant metal in the earth's crust, representing approximately 8% of total mineral components (Verstraeten, et al., 2008). Due to its reactivity, aluminum in nature is found only in combination with other elements such as sulphate, chloride etc.
Dietary aluminum is ubiquitous but in such small quantities that it is not a significant source of concern in persons with normal elimination capacity. Urban water supplies may contain a greater concentration because water is usually treated with aluminum before becoming part of the supply. Subsequent purification processes that remove organic compounds take away many of the same compounds that bind the element in its free state, further increasing aluminum concentration (Proudfoot,2009).

Animal studies in rats and case reports have implicated the use of oral aluminum-containing antacids during pregnancy as a possible cause for abnormal fetal neurologic development (Shuchang, et al., 2008; Gilbert-Barness, et al., 1998). Evidence for contribution of Aluminium to Alzheimer's disease remains contradictory (Flaten, 2001; Gupta, et al., 2005). Epidemiological studies have indicated a link between Aluminium in drinking water and Alzheimer's disease(AD) and a variety of human and animal studies have implicated learning and memory deficits after Aluminium exposure (Buraimoh *et al*.,2011a; Exley, 2005; Schmidt *et al*., 2001; Yokel, 2000).Aluminium Chloride was implicated to have negative effects on behavioural endpoints of wistar rats(i.e. alters behaviour), have negative effects on anxiety-related behaviour of wistar rats as it increased the rate of anxiety in aluminium treated rats and was also said to have neurodegenerative effects on the histology of cerebral cortex of adult wistar rats especially at higher dose (Buraimoh, et al., 2011b; Buraimoh, et al., 2011c; Buraimoh, et al., 2012d). This study was aimed at evaluating the histological effects that Aluminium Chloride Exposure could have on the Testis of Wistar Rats.

2. Materials and Methods

This study was conducted in the Department of Human Anatomy, Faculty of Medicine, Ahmadu Bello University, Zaria, Nigeria. The rules and regulations governing animal handling of Ahmadu Bello University were strictly adhered to and the experiment was conducted in accordance to the ethical committee guidelines.

Experimental animals: Ten male wistar rats were used for this experiment. The wistar rats were housed in steel cages maintained at good environmental conditions with sufficient food, water and under good ventilation. The wistar rats were kept for two weeks before commencement of Aluminium chloride administration. This was to enable the wistar rats acclimatized to the environment.

Experimental Design: The wistar rats were divided into five groups: control group I was given distil water while the four Aluminium exposed groups were given various concentrations of aluminium chloride as follows:
Group II received 475mg/Kg
Group III received 950mg/kg
Group IV received 1,425mg/kg
Group V received 1,900mg/kg.

The route of administration was through oral intubation and the duration of administration was Eight weeks. After eight weeks of administration, the wistar rats were humanely sacrificed and the testes were removed and immediately fixed in bouin's fluid. The testes were then transferred into an automatic tissue processor where they went through a process of dehydration in ascending grades of alcohol 70, 80, 95% and absolute alcohol for 2 changes each. The tissues were then cleared in Xylene and embedded in paraffin wax. Serial sections of 5 micron thick were obtained using a rotary microtome. The tissue sections were deparaffinised, hydrated and stained using the routine haematoxylin and eosin staining method (H&E). (Gurr, 1962). The stained sections were examined under the light microscope fitted to a laptop and digital camera for photomicrographs

3. Results and Discussion

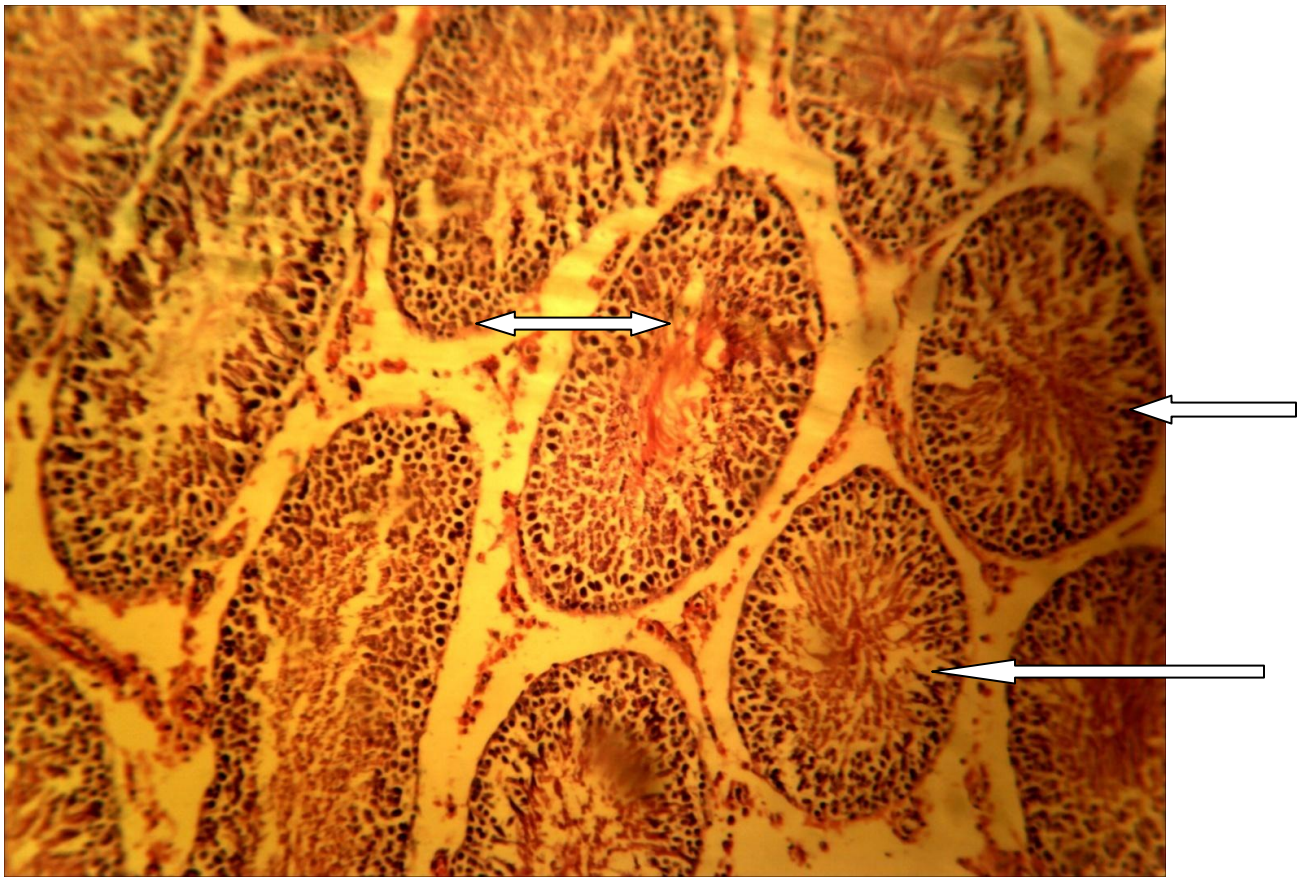

Fig.1. Photomicrograph of section in the normal testis of Control group I showing many seminiferous tubules (double arrow) with narrow lumina (arrows) and stratified epithelial lining .X100 H&E.

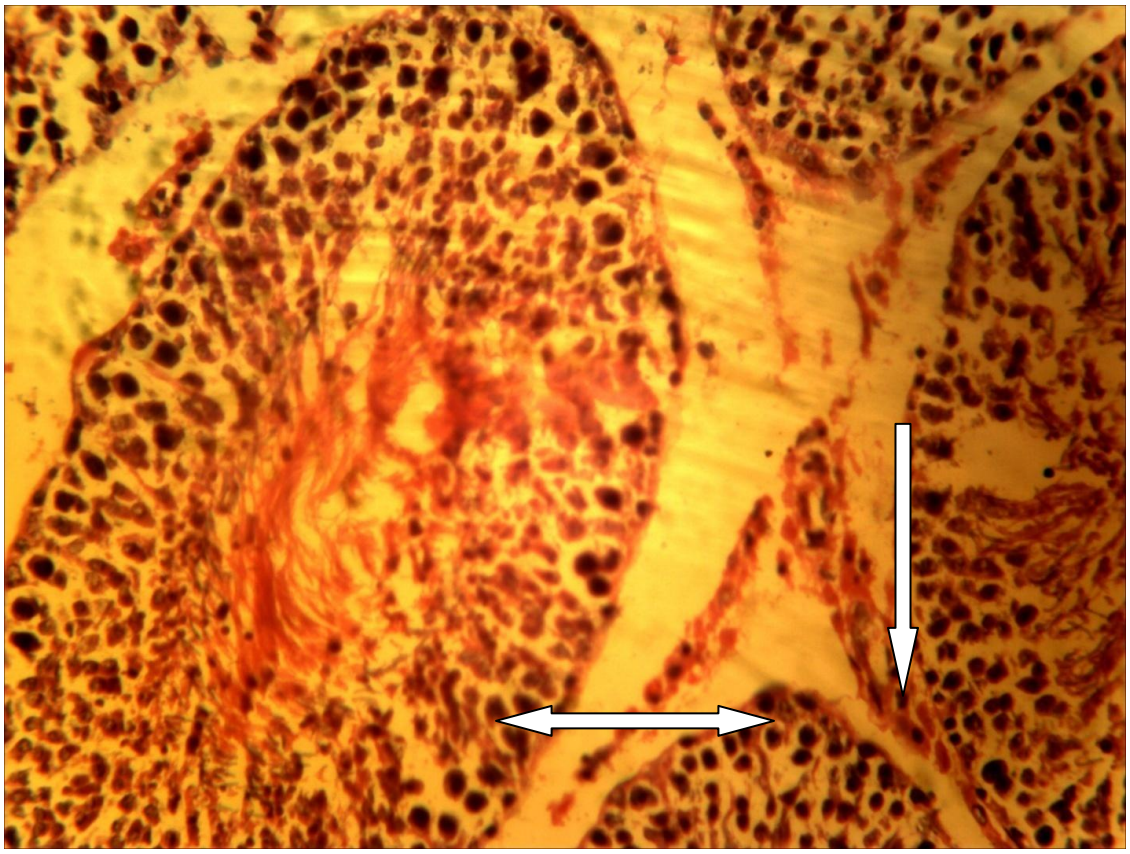

Fig.2. Photomicrograph of section in the normal testis of Control group I showing the seminiferous tubules are lined by spermatogonia(double arrow) The interstitium has small interstitial Leydig cells(arrow) .X250 H&E.

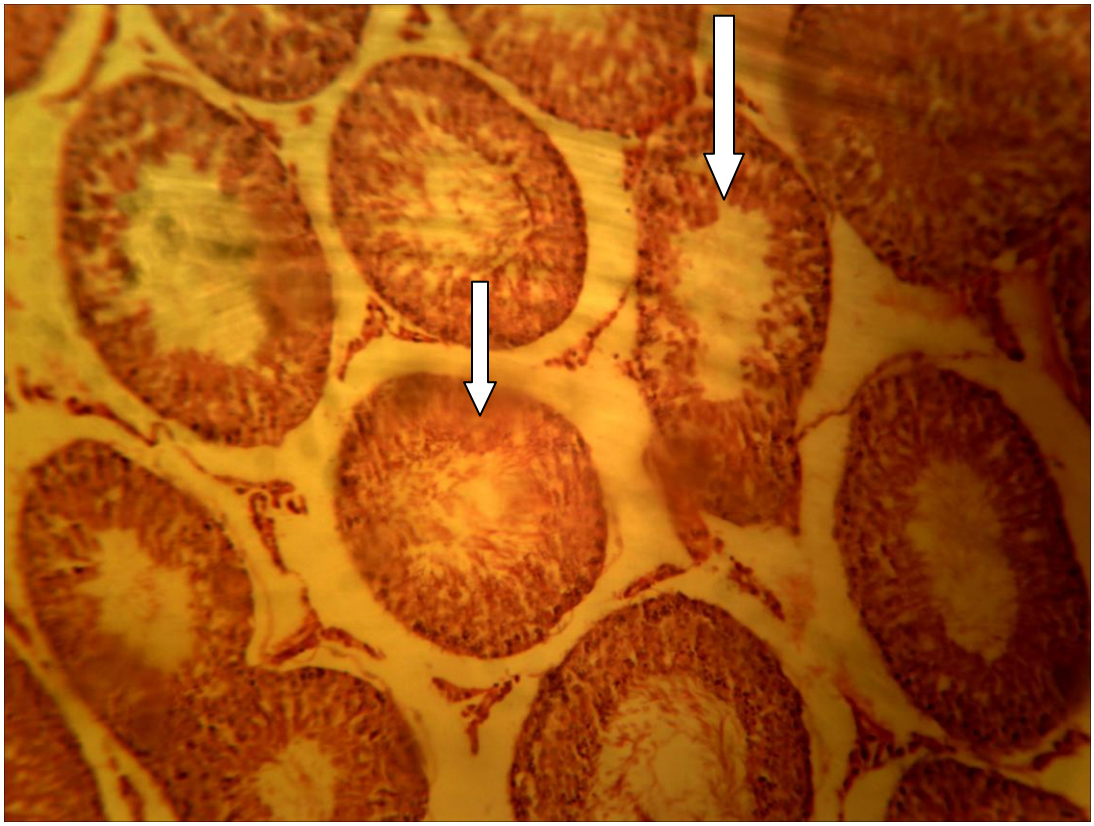

Fig.3. Photomicrograph of section in the testis of group II showing the seminiferous tubules which attained different shapes (arrows). X100 H&E

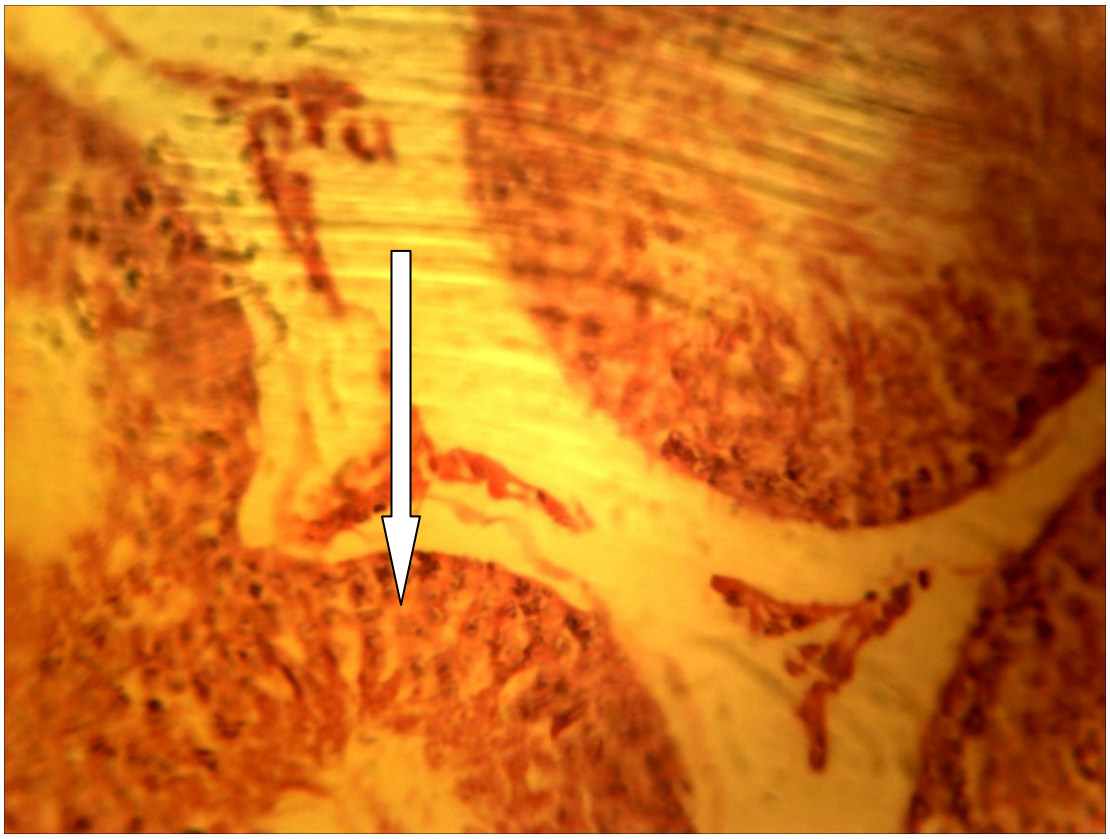

Fig.4. Photomicrograph of section in the testis of group II showing the seminiferous tubules that have lost the normal distribution of epithelial lining (arrow) .X250 H&E

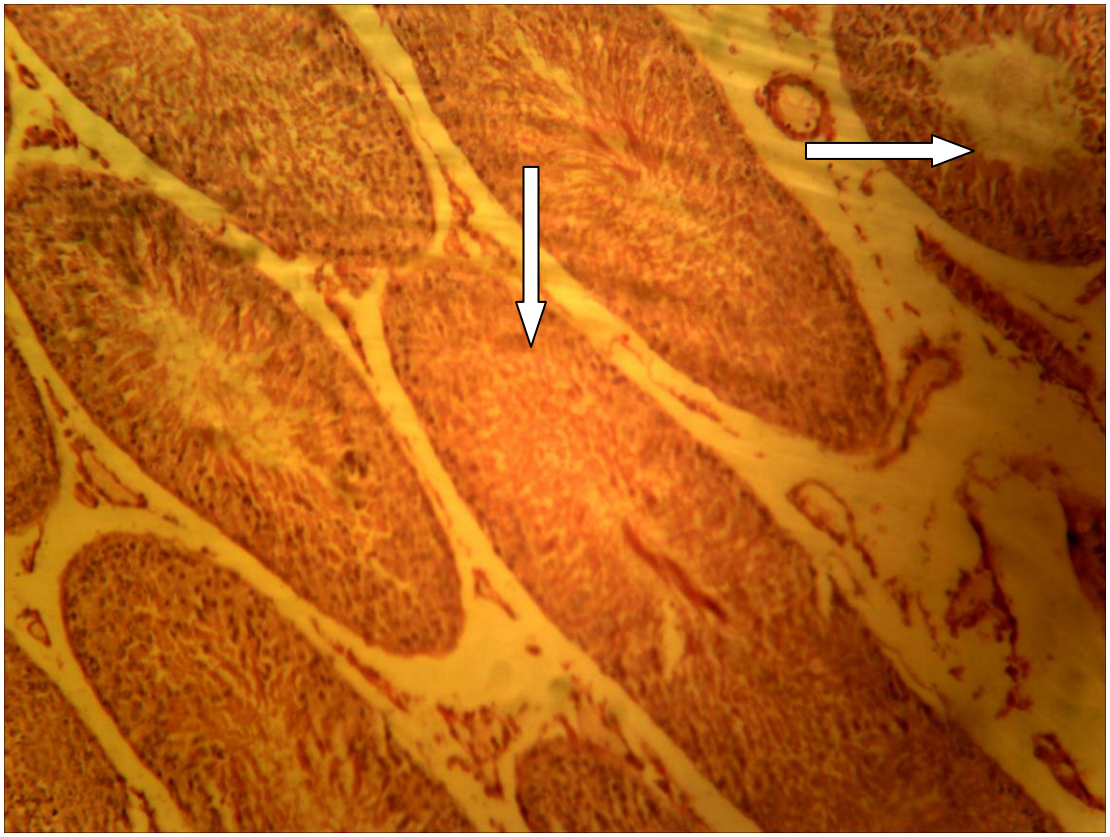

Fig. 5. Photomicrograph of section in the testis of group III showing the seminiferous tubules which attained different shapes (arrows). X100 H&E

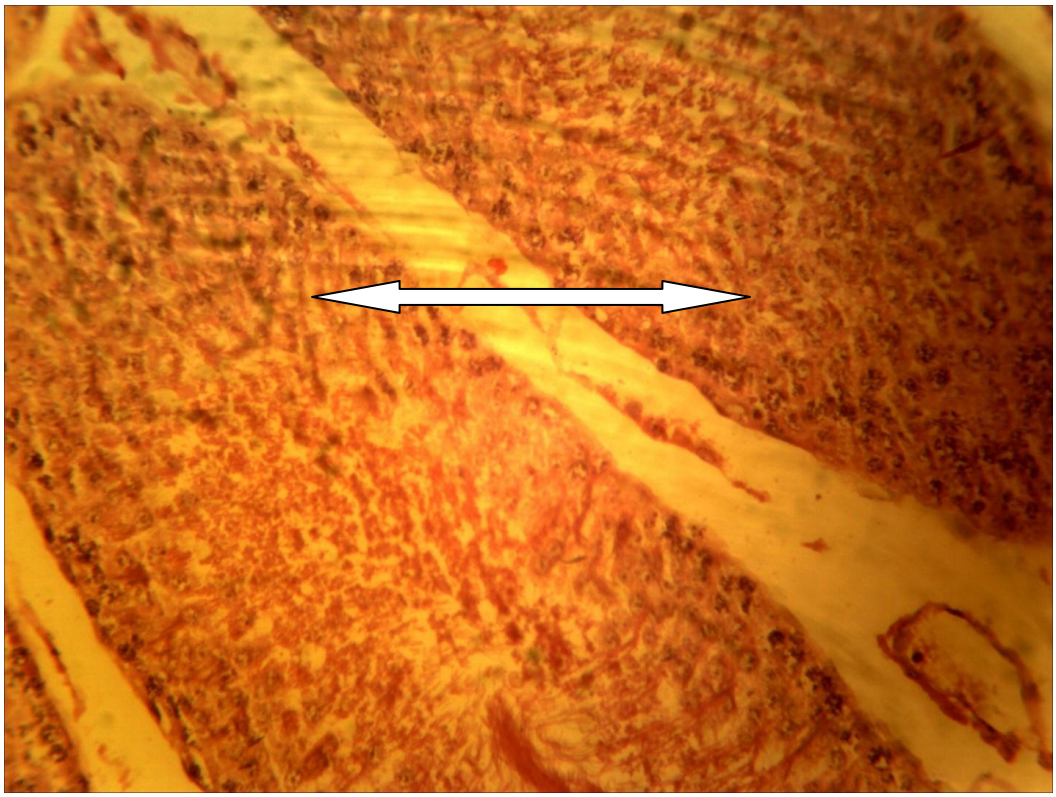

Fig.6 Photomicrograph of section in the testis of group III showing the seminiferous tubules that have lost the normal distribution of epithelial lining (double arrows) .X250 H&E

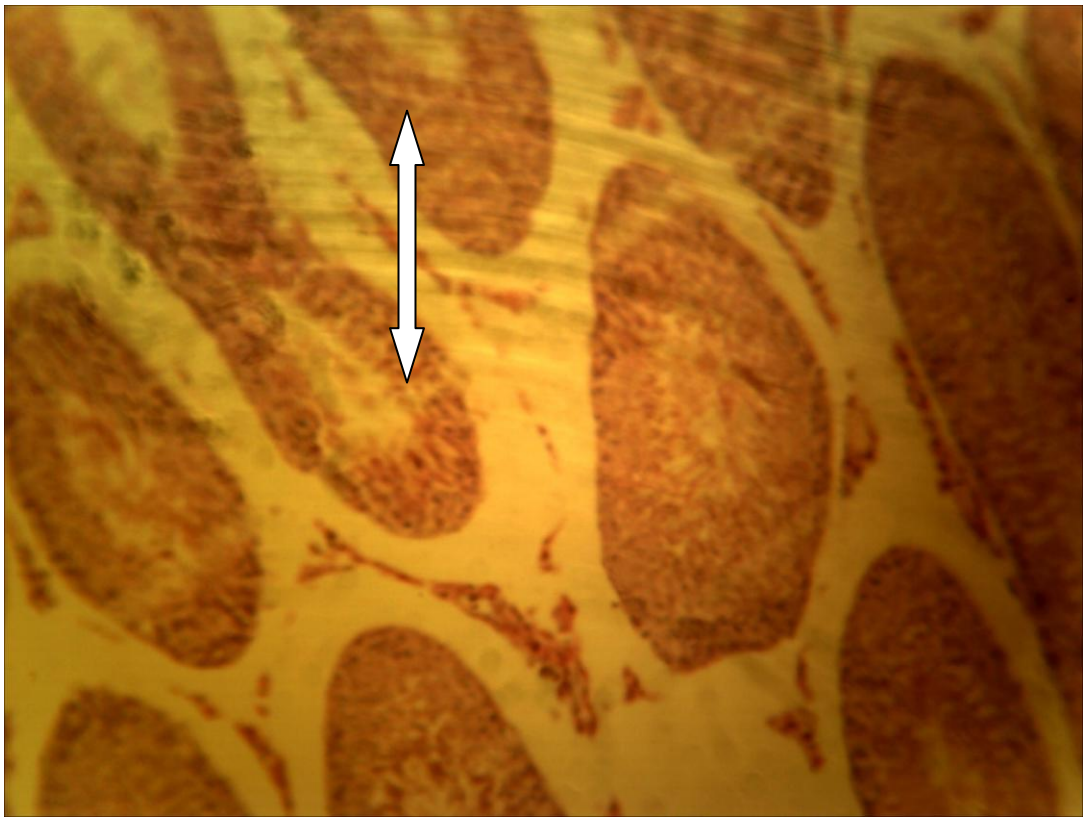

Fig.7 Photomicrograph of section in the testis of group IV showing distorted seminiferous tubules which attained different shapes (double arrow) .X100 H&E

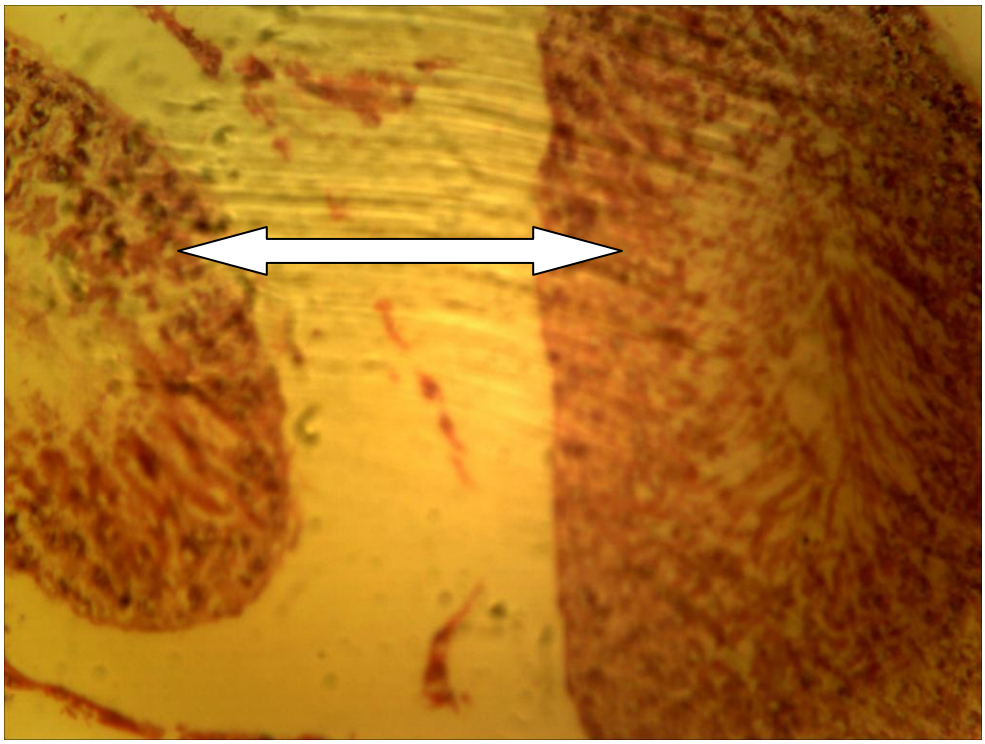

Fig.8 Photomicrograph of section in the testis of group IV showing distorted seminiferous tubules which attained different shapes (double arrow) .X250

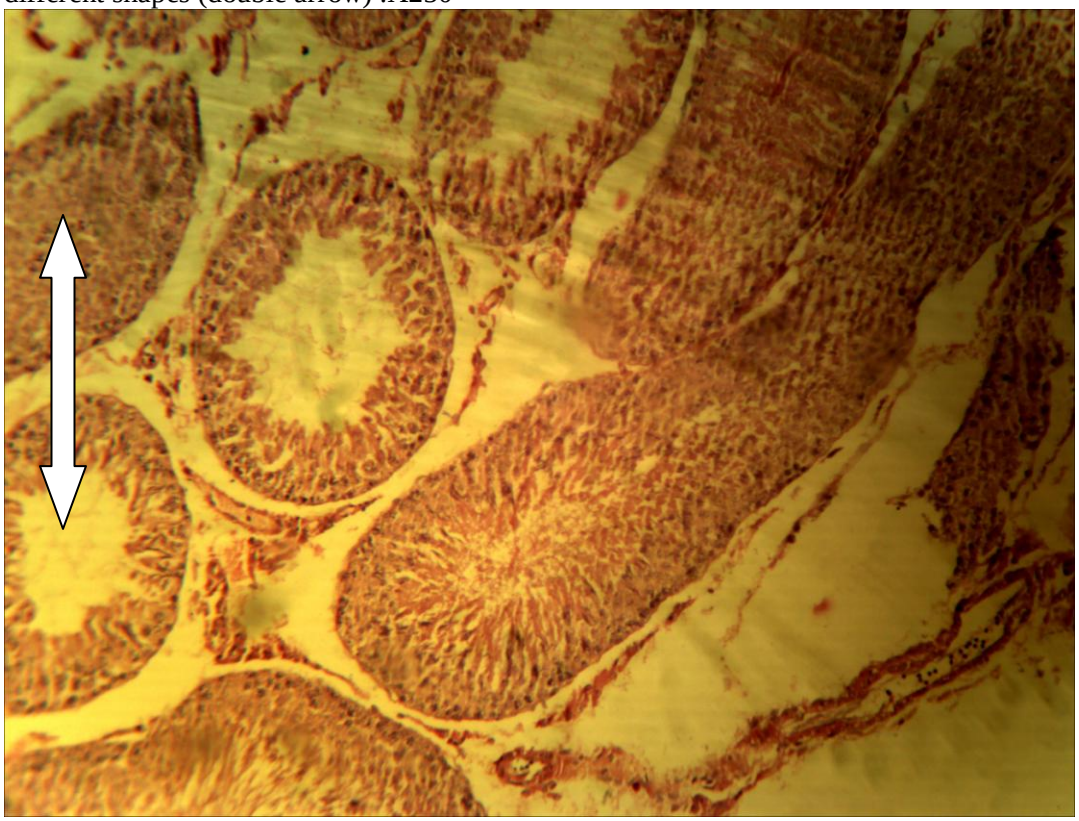

Fig.9 Photomicrograph of section in the testis of group V showing distorted seminiferous tubules with loss of normal distribution of epithelial linning and vacuolar cytoplasm (double arrow)

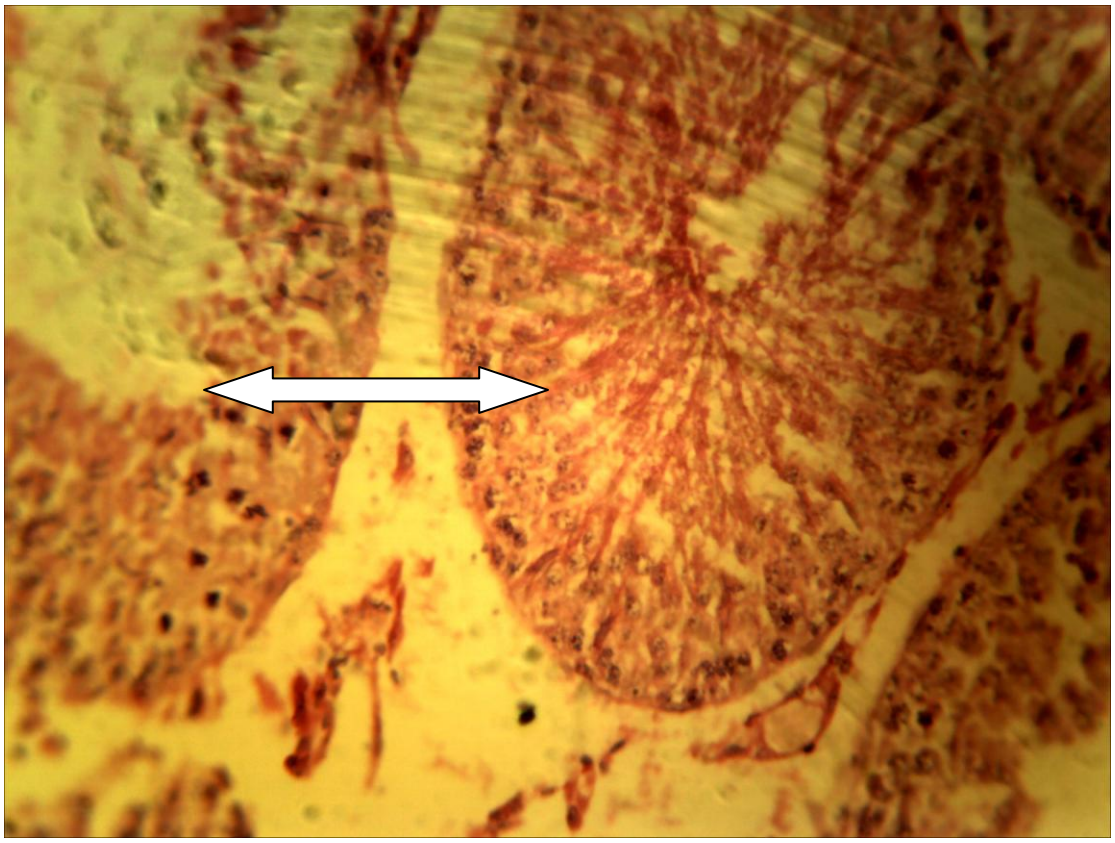

Fig10. Photomicrograph of section in the testis of group V showing distorted seminiferous tubules with loss of normal distribution of epithelial linning and vacuolar cytoplasm (double arrow).

The testes are responsible for making testosterone, the primary male sex hormone, and for generating sperm. Within the testes are coiled masses of tubes called seminiferous tubules. These tubules are responsible for producing the sperm cells through a process called spermatogenesis. (Elaine, 2004; Scott,2000).

Aluminum has the potential to be neurotoxic in human and animals. Although, aluminum has been implicated in Alzheimer's disease, Parkinsonism, Dementia complex and causes extensive damage to the nervous system, to date the mechanism of Aluminium neurotoxicity has not been fully elucidated (Niu, *et al*., 2007). It presents in many manufactured foods and medicines and is also added to drinking water for purification purposes (Newairy, *et al*., 2009).

The results for wistar rats in group I (control), where only distil water were administered showed normal histological features of the testis as indicated by well arranged seminiferous tubules lined by spermatogonia (Fig.1&2). The results of group II that received 475mg kg^{-1} of aluminium chloride showed seminiferous tubules that attained different shapes and have lost the normal distribution of epithelial lining (Fig.3&4). For the wistar rats in group III and IV that received 950mg kg^{-1} and 1425mg kg^{-1} of aluminium chloride respectively showed seminiferous tubules that attained different shapes and have lost the normal distribution of epithelial lining (Fig.5-8). The wistar rats in group V that was administered with 1900mg kg^{-1} of aluminium chloride showed marked distorted seminiferous tubules with loss of normal distribution of epithelial linning and vacuolar cytoplasm(Fig.9&10) which are indications of testes degeneration that could affect sperm cells production..

Our observations revealed that the wistar rats in group I that received distil water showed normal architecture of the histology of the testes while the aluminium treated groups(II-V) showed some degree of degenerations of the testes (Fig.3-10) when compared with the control(Fig.1&2).

4. Conclusion

Based on our observation, we therefore conclude that aluminium chloride exposure could be detrimental to the integrity of the testis of wistar rats.

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References

Buraimoh, A.A., Ojo, S.A., Hambolu, J.O., & Adebisi, S.S. (2011a). Effects of oral administration of aluminium chloride on the histology of the hippocampus of wistar rats. Curr. Res. J. Biol. Sci.,3: 509-515.

Buraimoh, A.A., Ojo, S.A., Hambolu, J.O., & Adebisi, S.S. (2011b). Behavioural enpoints of adult wistar rats, following aluminium chloride exposure. Br. J. Pharmacol. Toxicol., 2: 273-276. ISSN: 2044-2467.

Buraimoh, A.A., Ojo, S.A., Hambolu, J.O., & Adebisi, S.S. (2011c). Effects of Aluminium Chloride on Anxiety-Related Behaviour. American Journal of Neuroscience 2(2): 65-69, 2011. ISSN 1948-9900. Science Publications.

Buraimoh, A.A., Ojo, S.A., Hambolu, J.O., & Adebisi, S.S. (2012d). Effects of Aluminium Chloride Exposure on the Histology of the Cerebral Cortex of Adult Wistar Rats. Journal of Biology and Life Science. Vol.3, No.1. ISSN 2157-6076. DOI: 10.5296/jbls.v3i1.1421. @Macrothink Institute.

Elaine, N.M. (2004). Human Anatomy and Physiology. 6th Edn., Retrieved from: http://www.ehow.com/facts-5521847-function-anatomy-human-testes.html# ixzz1 VsszSqbg.

Exley, C. (2005). The aluminium-amyloid cascade hypothesis and Alzheimer's disease aluminium and β-amyloid. Alzheimer's Disease, 38: 225-234. DOI: 10.1007/0-387-23226-5_11.

Flaten, T.P. (2001). Aluminium as a risk factor in Alzheimer's disease, with emphasis on drinking water. Brain Res. Bull., 55: 187-196. DOI: 10.1016/S0361-9230(01)00459-2.

Gilbert-Barness, E., Barness, L.A., Wolff, J., & Harding, C. (1998). Aluminum toxicity. *Arch Pediatr Adolesc Med*. May 1998; 152(5):511-2.(Medline).

Gupta, V.B., Anitha, S., Hegda, M.L., Zecca, L. & Garruto, R.M., et *al*. (2005). Aluminium in Alzheimer's disease: Are we still at a crossroad? Cell. Mol. Life Sc., 62: 143-158. DOI: 10.1007/s00018-004- 4317-3.

Gurr, E. (1962). Staining Animal Tissues, Practical and Theoretical Leoned Hill, London.

Gray, H. (2000). Splanchnology. The Male Genital. In: *Anatomy of the Human Body*. 20th ed. Philadelphia, New York: Lea & Febiger, 1918; Bartleby 2000; 2000.

Jiang, H.X., Chen, L.S., Zheng, J.G., Han, S., Tang, N., & Smith, B.R. (2008). Aluminum-induced effects on Photosystem II photochemistry in citrus leaves assessed by the chlorophyll a fluorescence transient. *Tree Physiol*. Dec 2008; 28(12):1863-71. (Medline).

Michael, H.R., & Wojciech, P. (2006). Histology Text and Atlas. 5th Edn., Lippincott Williams & Wilkins.

Mosby's Medical Dictionary, 8th edition. © 2009, Elsevier**.**

Newairy, A.S., Salama, A.F., Hussien, H.M., & Yousef, M.I. (2009). Propolis alleviates aluminium-induced lipid peroxidaion and biochemical parameters in male rats. Food Chem Toxicol. 47(6):1093-8.

Niu,Q., Yang, Y., Zhang, Q., Niu, P., He, S., Di Gioacchino, M., & Boscolo P. (2007). "The relationship between Bcl-2 gene expression and learning & memory impairment in chronic aluminum-exposed rats. Volume 12, Number 3, 163-169, DOI: 10.1007/BF03033913.

Proudfoot, A.T. (2009). Aluminium and zinc phosphide poisoning. *Clin Toxicol (Phila)*. Feb 2009; 47(2):89-100. (Medline).

Romer, A.S., & Parsons, T.S. (1977). The Vertebrate Body. Philadelphia, Holt-Saunders International, PA,pp: 385-386.

Schmidt, M.L., Zhukavera, V., Perl, D.P., Sheridan, S.K., Schuck, T., *et al*.(2001). Spinal cordneurofibrillary pathology in alzheimer disease and guam parkinsonism-dementia complex. J. Neuropathol. Exp. Neurol., 60: 1075-1086.

Scott, F.G. (2000). Developmental Biology Online Textbook, 6th Edn. Sinauer Associates, Inc. of Sunderland (MA).

Shuchang, H., Qiao, N., & Piye, N. (2008). Protective effects of gastrodia elata on aluminium-chloride-induced learning impairments and alterations of amino acid neurotransmitter release in adult rats. *Restor Neurol Neurosci*. 2008;26(6):467-73. (Medline)

Skinner, M., McLachlan, R., & Bremner, W. (1989). Stimulation of sertoli cell inhibin secretion by the testicular paracrine factor PModS. Mol. Cell. Endocrinol., 66(2): 239-249. Doi: 10.1016/0303- 7207(89)90036-1.

Verstraeten, S.V., Aimo, L., & Oteiza, P.I. (2008). Aluminium and lead: molecular mechanisms of brain toxicity. *Arch Toxicol*. Nov 2008; 82(11):789-802.(Medline).

Yokel, R.A. (2000).The toxicology of Aluminium in brain: review, Neurotoxicology.21 (5):813-828.