

Reldan and Ammonium Nitrate Induced Histopathological and Fine Structural Changes in Testis of Mice

Mahmoud, A. El-Banhawy¹, Nahed, H.A. Riad², Shadia, A. Radwan³, Mona El-Akkad⁴ and Aziza El-Wessemy⁵

^{1, 2, 4}Zoology Depts, Faculty Science, ^{3, 5}Biology & Geology Depts, Faculty of Education, Ain Shams University

Abstract

Two agrochemicals, reldan insecticide and ammonium nitrate fertilizer affected the histopathological and ultrastructural of mice testicular tissue. Mice were divided into 13 groups, the first group served as a control group while the other groups treated with reldan or ammonium nitrate. For each of the agrochemicals used, the $\frac{1}{2}$ LD₅₀ (12 mg/kg b.wt. for reldan and 90 mg/kg b.wt. for ammonium nitrate) was given as a single dose to 3 groups of mice which were sacrificed after 5, 10 and 15 days post-treatment, and as repeated daily doses to other groups of mice for 5 and 10 days. Animals treated for 5 days were sacrificed 24 hrs after the last dose, while those treated for 10 days were sacrificed 24 hrs or 5 days after the last dose. The applied agrochemicals caused histopathological changes in the testes including, disorganization of the basal lamina and germinal epithelium, maturation arrest at various degrees of degenerated spermatocytes and spermatids in the testicular tissue. Ultrastructural examination showed marked alterations and degeneration of organelles in germ cells, Sertoli cells and Leydig cells.

Introduction

Although, these agrochemicals (fertilizers and insecticides) were planned to increase the food resources and improve both quality and quantity of the beneficial crops besides protecting them against the attacks of pests, they also act as potential hazards for health in general. *WHO (1975), Dunnik et al. (1984) and WHO (1992)* recorded serious unwanted responses affecting man and his domestic animals, after the utilization of agrochemicals. Reldan insecticides, the choice in the present investigation, is one of the wellknown organophosphorus compounds. *WHO (1992)* reported that the toxicity of organophosphorus compounds is by phosphorylation of acetylcholine esterase, accumulation of acetylcholine at the nerve synapses causing serious nervous disorders. Observations of *Ray et al. (1988, 1992)* showed that the testes of rats undergo histopathologic changes when treated with the organophosphorus compound "quinalphos" (250 mg/kg b.wt.) after 13 and 26 days. These

changes include shrinkage of the seminiferous tubules, degeneration of the germinal epithelium and appearance of large number of Sertoli cells indicating testicular atrophy. Thus, they concluded massive degeneration of all varieties of germ cells, and reduction in the spermatozoa count. Also, the testes of albino mice undergo histopathologic changes when treated with organophosphorus "dimethoate". These changes included alterations of the normal architecture of tubules, disorganization with variable degrees of degeneration (hypoplasia and spermatogenic arrest), as well as necrosis of cells especially spermatocytes and spermatids. In addition, hemorrhage and reduction of tubular diameter were reported (*Sanad, 1993; Yosef, 1993; El-Sayyad et al., 1995; Akbarsha and Sivasamy, 1997; Debnath and Mandal, 2000; Shaalan et al., 2002 and Ismail, 2005*).

The toxic effects of ammonium nitrate have been described by *Mohamed, (1993)* who recorded marked degenerative changes

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in the liver of rats. Histological examination of testes, after excessive application of the fertilizer superphosphate, revealed degenerative changes including atrophy of the seminiferous tubules, hyperplasia of the germinal epithelium and thickening of the walls of blood vessels (Ray *et al.*, 1997). On the other hand, the toxicity of ammonia was followed by lack of oxygen in tissue and organs (hypoxia) which led to a decrease in the metabolic rate of the organs (Saeed, 1997).

According to El-Akkad and Abou Dief (1999) many alterations in the ileal mucosa of the cattle egret, treated with ammonium nitrate fertilizer were observed. The degenerative changes included vacuolation and weaker stainability of the cytoplasm, the signs of degeneration and swollen mitochondria with completely degenerated cristae were observed. The rough endoplasmic reticulum (RER) revealed degranulation of its cisternae. The detached ribosomes were scattered forming clusters of polyribosomes. The cisternae of smooth endoplasmic reticulum (SER) were increased.

Material & Methods

Experimental Animals :

Sixty-five male albino mice, *Mus musculus* were used in the present investigation. All experimental animals were fed standard diet and kept under suitable conditions during the whole period of experimentation. The mice were 3 ± 1 months old with an average weight of 25 ± 5 g.

The Applied Agrochemicals

In the present work, the insecticide, reldan was obtained from Ministry of Agriculture, Egypt. The 24 hours LD₅₀ of this compound was determined according to Reish and Oshida (1986) as 24 mg/kg.b.wt. for mice. On the other hand, the fertilizer "ammonium nitrate" was purchased from El-Nasr Company for Coke and Basic Industry, Cairo, Egypt. The 24 hours LD₅₀ of this compound was determined according to Reish and Oshida (1986) as 180mg/kg.b.wt for mice.

Experimental Design

Mice were divided into 13 groups (M1-M13), 5 animals per group as follows:

Group (M1) : Animals were kept without treatment and served as a control group.

Group (M2) : Each animal received a single oral dose of the $\frac{1}{2}$ LD₅₀ of reldan (12mg/kg.b.wt.) and specimens were obtained after 10 days.

Group (M3) : Each animal received a single oral dose of the $\frac{1}{2}$ LD₅₀ of reldan (12mg/kg.b.wt.) and specimens were obtained after 10 days.

Group (M4) : Each animal of this group received a single oral dose of 12 mg/kg.b.wt. of reldan and specimens were sacrificed after 15 days.

Group (M5) : Each mouse of this group received a daily oral dose of reldan (2.4 mg/kg.b.wt.) for 5 successive days and the specimens were sacrificed at the end of the treatment.

Group (M6) : Each mouse of this group received a daily oral dose of reldan (1.2 mg/kg.b.wt.) for 10 successive days and the specimens were sacrificed at the end of the treatment.

Group (M7) : Each mouse of this group received a daily oral dose of reldan (1.2 mg/kg.b.wt.) for 10 successive days then the specimens were sacrificed on the 15th day.

Group (M8) : Each animal of this group was given a single oral dose of $\frac{1}{2}$ LD₅₀ of fertilizer (90mg/kg.b.wt.) and the specimens were sacrificed after 5 days.

Group (M9) : Each mouse of this group received a single oral dose of $\frac{1}{2}$ LD₅₀ of the fertilizer (90mg/kg.b.wt.) and the specimens were sacrificed after 10 days.

Group (M10) : Each mouse of this group received a single oral dose of the fertilizer (90mg/kg.b.wt.) and the specimens were sacrificed after 15 days.

Group (M11) : Each mouse of this group received a daily oral dose of the fertilizer (18mg/kg.b.wt.) for 5 successive days and the specimens were sacrificed at the end of the treatment.

Group (M12) : Each mouse of this group received a daily oral dose of the fertilizer (9mg/kg.b.wt.) for 10 successive days and

the specimens were sacrificed at the end of the treatment.

Group (M13) : Each mouse of this group received a daily oral dose of the fertilizer (9mg/kg.b.wt.) for 10 successive days, then the specimens were sacrificed on the 15th day.

1.Histological preparations

Specimens used for paraffin sectioning were fixed in Bouin's fluid for 24 hours, washed in 10% alcohol containing few drops of saturated lithium carbonate to remove the excess of picric acid, dehydrated in ascending series of ethyl alcohol, followed by clearing in terpineol. Then, the specimens were immersed immediately in 3 changes of benzol before embedding in paraffin wax. Sections of 5 microns thickness were mounted on clean glass slides without using any adhesive fluid. The mounted sections were stained with Mayer's haematoxylin and eosin.

2.Electron microscopic preparations

For the ultrastructural purposes, small pieces (about 1mm³ in size) of the testicular tissue were immediately fixed in 2.5% glutaraldehyde in 0.1 M phosphate buffer at pH 7.4 at room temperature for 4 hours, then rinsed twice in 0.1 M phosphate buffer (15 min in each). Specimens were post fixed in 2.0% buffered osmium tetroxide for half an hour at 4°C. The tissue specimens were then washed twice in phosphate buffer for 30 min, then dehydrated in ascending grades of ethyl alcohol for two changes, 15 minutes in each grade. Specimens were then cleared in propylene oxide for two changes 15 min each and infiltrated at room temperature in a mixture of spurr and propylene oxide. Then, in pure spurr for 3 hours or even over night. Using been capsules, specimens were embedded in spurr. Semi-thin sections of μm thickness were cut, picked up on glass slides, stained by toluidine blue and examined for general orientation with the light microscope. Specimens were then retrimmed to the selected region and ultra-thin sections, 60-nm thickness were cut and picked up on copper grids. Sections

mounted on grids were double stained using uranyl acetate and lead citrate. Staining was done in the dark for 20 min. Grids were washed in three successive glass bottles containing distilled water. After the last bath, grids were dried on a filter paper. Stained grids were examined and photographed by a Joel 1200EX 2 transmission electron microscope, at the Faculty of Science, Ain Shams University.

Results and Discussion

The testis has been selected in the present investigation to discern and evaluate any possible impacts of the two of the widely used agrochemicals in Egypt, namely the isecticide reldan and the fertilizer ammonium nitrate. The histological appearance of the control testis (P1.1, Figs. 1-3) reveals spermatogonia which lie next to the basal lamina and have two types, the A type characterized by a large oval nucleus with condensed chromatin and the B type characterized by centrally located nuclei having dispersed chromatin. Spermatogonia type B cells differentiate into primary spermatocytes (PL 1, Fig 3). They are the largest germ cells seen in the section of seminiferous tubules and disposed in two or three layers. Spermatids are formed from secondary spermatocytes. The early spermatids are distinguished by their small size, rounded nuclei with condensed chromatin. In the late spermatide, the nucleus became more ovoid with different stainability (PL.1, Figs. 2 & 3), these spermatids are arranged with their acrosomes toward the Sertoli cells (PL.1, Fig. 2). The spermatids differentiate to spermatozoa (through process known as spermiogenesis) which are found in clusters in the lumina of the seminiferous tubules. Sertoli cells are commonly referred to as sustaining, nutritive or nurse cells, they are elongated cells with oval nuclei. The heads of the late spermatids are attached to Sertoli cells while their tails lie free within the lumens (PL.1, Fig. 3). The spaces between the seminiferous tubules in the testis, interstitial tissues, are filled with connective tissue, nerves, blood and lymphatic vessels and

Leydig cells, which are found in clusters placed close to the blood capillaries. Leydig cells are relatively large in size, polyhedral in shape and possessed a large spherical nuclei with dispersed chromatin, and one or two nucleoli (PL.1, Fig. 3).

Reldan treated-Mice

The histological description of the testes of reldan-treated mice (groups M2-M7) revealed various degrees of degenerative changes. The histological observations of the testes of reldan-treated mice (group M2) revealed marked disruption in the basal lamina of some seminiferous tubules leading to disorganized germinal lineage and disturbance of the general architecture (PL.1, Figs 4-6). The spermatogenic cells were reduced (hypoplasia) with maturation arrest at the spermatid stages associated with a decrease in the number of sperms. Multinucleated giant cells, exfoliation of spermatid cells within the lamina of some tubules, sloughing in other seminiferous tubules and hemorrhagic areas within the interstitial spaces were also observed (PL.1, Figs 5 & 6). The testes of mice of group M3 revealed general impairment of the seminiferous tubules (PL.2, Figs. 7 & 8). Most of the seminiferous tubules showed malformed irregular shapes and disruption of the basal lamina in some places. Cytoplasmic degeneration of germinal epithelium (vacuoles), hypoplasia and maturation arrest at spermatid cells with sloughing of germ cells were also seen within the lamina of some tubules (PL. 2, Figs. 7 & 8). The testes of mice of group M4 revealed that some of the seminiferous tubules exhibited improvement (to a certain limit) with slight changes included appearance of the different stages of spermatogenesis with an increase in the number of spermatozoa in some tubules. Whereas slight histopathological alterations represented by maturation arrest at spermatid stages, congestion and hemorrhage of blood vessels were noticed (PL.2, Figs. 9 & 10). The testes of mice treated with repeated doses of reldan (group M5) showed some damages which were mainly of the mild type (PL. 2, Figs. 11 & 12). Some seminiferous tubules appeared normal with

almost the frequent series of spermatogenesis. The basal lamina of the affected seminiferous tubules revealed malformed irregular shapes and disruption in some places (Fig. 12). The spermatogonia and some primary spermatocytes became necrotic, degenerated and their nuclei exhibited pyknosis (Fig. 11). In the intertubular spaces, patches of inflammatory cells were clearly seen. The spaces exhibiting distinct hemorrhage and the blood vessels displayed, marked congestion (PL. 2, Figs. 11 & 12). *Balasubramanian et al. (1980)* explained the hyperemic condition (congestion) of blood vessels to the inhibition of prostaglandin's synthesis, since these compounds are known to regulate the testicular blood flow of rats. Furthermore, *Singwi and Lall, (1980)* assumed that increased breakage of blood capillaries lead to further augmentation of interstitial oedema and consequent disorganization of effect of Leydig cells in the interstitial tissue of mammalian testes. According to *Haschek and Rousseaux (1991)*, lesions of testes occurred as a result of inflammatory reaction in the interstitial tissues. The interstitial macrophage cells may increase and phagocytize any necrotic debris present in the interstitial space, thus causing interstitial oedema and finally cell death within the tubules. Thus, it is likely that congestion of blood vessels and hemorrhage in the testes of treated animals, presented here, might be due to inflammation reactions in the interstitial tissues.

Treatment with the repeated doses of reldan for 10 successive days (group M6) induced serious damage in most of the seminiferous tubules in the testes of mice (PL.3, Figs. 13 & 14). The basal lamina of the majority of tubules was limited by fibrotic deeply stained connective tissue. Various degrees of spermatogenic inhibition and completely arrested spermiogenesis were clearly noticed in the seminiferous tubules. Besides, cytoplasmic degeneration in primary spermatocytes, multinucleated giant cells with cytoplasmic debris were observed (PL.3, Figs. 13 & 14). Earlier experiments suggested that eosinophilic masses in the lumens of seminiferous tubules of rats represented testicular

necrotic debris (*Singh and Chakravarti, 1968*). The testes of the group M7 showed slight regeneration of the germinal epithelium (PL.3, Figs. 15 & 16). This was represented by appearance of spermatogenesis process in some tubules. Other tubules exhibited some pathological changes as the presence of exfoliated germ cells and the appearance of congestion in some vessels (PL.3, Figs. 15 & 16).

Ammonium nitrate treated-Mice

Treatment with a single dose of ammonium nitrate fertilizer for 5 days (group M8) induced slight damage in most of seminiferous tubules in testes (PL.3, Figs. 17 & 18). The basal lamina of seminiferous tubules exhibited disruption at some places. The most important changes appeared as degeneration of the cytoplasm, pyknosis of the nuclei of spermatogonia and marked atrophy of primary spermatocytes. Maturation arrest of spermatide beside multinucleated giant cells were also detected within the lumen. Congestion of blood vessels and inflammatory cells with wide hemorrhagic areas were clearly seen (PL.3, Figs. 17 & 18). The testes of mice treated with a single dose of ammonium nitrate for 10 days (group M9) showed severe damage in most of the seminiferous tubules, irregular shape of basal lamina with disruption of some places and fusion of their tubular contents (PL.4, Figs. 19 & 20). Also, the epithelial germinal layers were separated from the basal lamina of some tubules & degeneration and atrophy of some spermatogenic cells with pyknotic features of their nuclei were detected (Fig. 20). In group M10, some seminiferous tubules appeared normal in shape and spermatogenic layers were comparatively well organized and developed (PL.4, Figs. 21 & 22). On the other hand, some tubules showed maturation arrest at spermatid stages, slight pyknosis of some spermatogenic cells and remarkable hemorrhage with blood vessels congestion in the interstitial spaces (PL.4, Fig. 22). The testes of mice treated with repeated doses of ammonium nitrate for 5 days (group M11) revealed mild degenerative alterations (PL.4, Figs. 23 & 24). The basal lamina

revealed discontinuity in some places and most spermatogonial cells showed necrotic features with pyknotic nuclei. Besides, abnormal multinucleated giant cells, spermatid arrested cells and cytoplasmic debris within the lamina of the tubules were observed (PL.4, Figs. 23 & 24). The testes of mice (group M12) exhibited severe damage in most of the seminiferous tubules such as disorganization and destruction of the normal architecture of germinal epithelium. The basal lamina became deeply stained and thickened, some tubules showed sloughing appearance and maturation arrest of spermatid stage reflected by reduction in the number of sperms (PL.5, Figs. 25 & 26). In mice of group M13, some tubules showed improvement (to a certain limit), the spermatogenic cells have restored their regular shape with the appearance of some sperms in the lumina of the tubules (PL.5, Figs. 27 & 28). On the other hand, signs of pyknotic nuclei of spermatogonia, maturation arrest hemorrhage in the inter tubular spaces were clearly seen in plate (5), Fig. (28). This study proved that agrochemicals (fertilizers) exert harmful effects on reproduction and cause profound histopathological changes in gonadal tissue (*Moutschen, 1985*).

Ultrastructure of the testis of mice

Electron microscopic examination of the testes of control mice (PL.5 & 6, Figs. 29-33) revealed that each seminiferous tubule is surrounded by a thin basal lamina and covered externally by a specialized zone of fibrous tissue (PL.5, Fig. 29). Seminiferous tubules are lined by a stratified epithelium which consists of spermatogenic and supporting Sertoli cells. These supporting or sustentacular nurse cells (Sertoli) are irregularly columnar in shape and directly attached to the basal lamina of the seminiferous tubule. The lateral and apical portions of Sertoli cells extend and reach the tubular lumen, giving out a sheetlike cell processes termed apical or lateral processes that surround the germ cells (PL.5, Fig. 30). The spermatogenic cells go through several cycles of development, involving cellular division and maturations. The spermatogonia are found

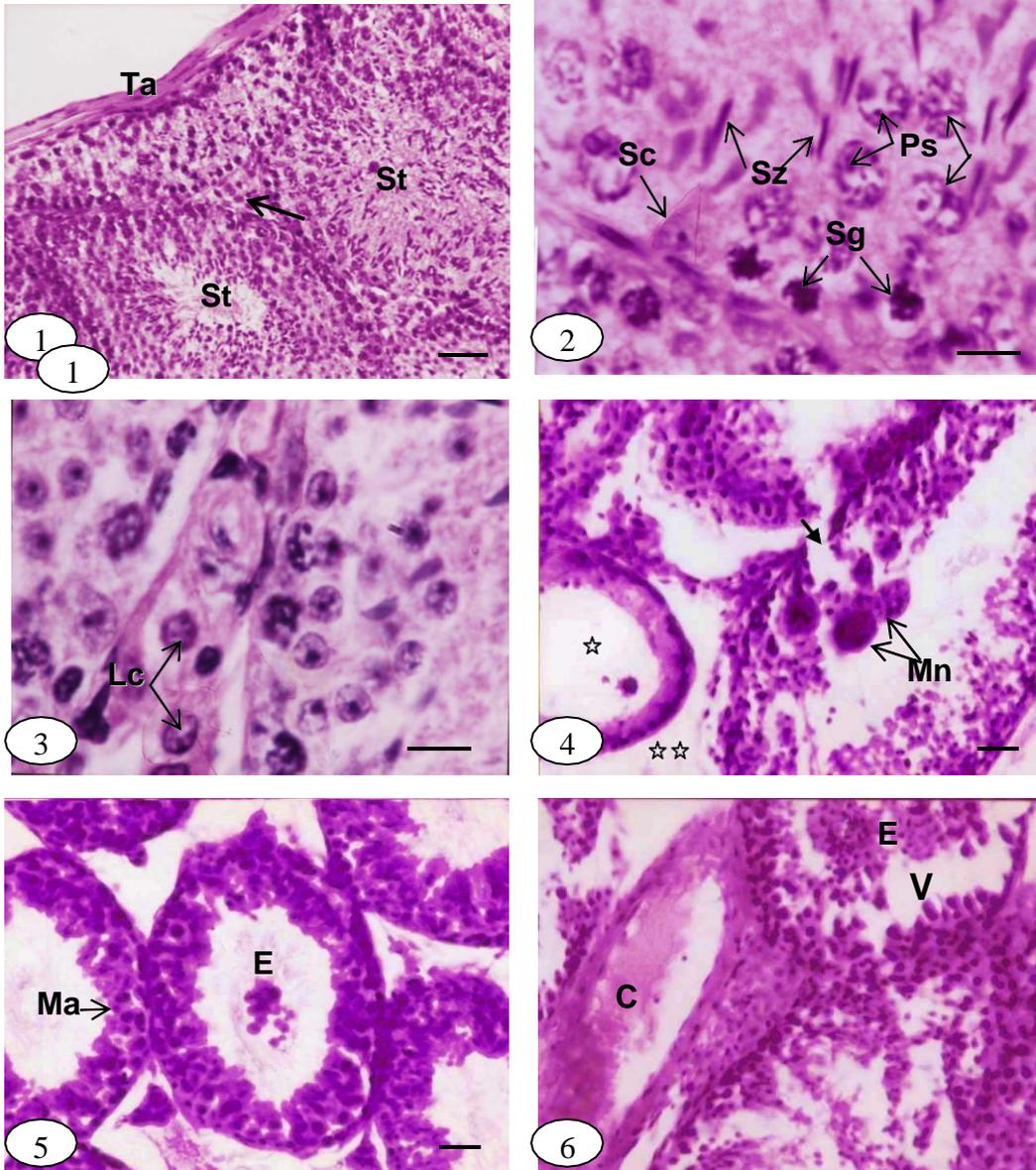
adjacent to the basal lamina and have two types A and B. Type B Spermatogonia are numerous than type A and have rounded nuclei with heavily stained heterochromatin masses attached to the nuclear membrane (Fig. 31). Mitochondria, rough endoplasmic reticulum and free ribosomes are easily observed in the cytoplasm (Figs. 31 & 32). The meiotic division of the spermatocytes results in the formation of the haploid spermatids. The spermatids are small rounded cells with spherical nuclei (PL. 6, Fig. 32). Spermatid development is conveniently divided into a number of phases: Golgi, cap, acrosome, and maturation phases, (Fig. 33). Leydig cells are the major cell type of the interstitial tissue, they are relatively large in size, ovoid or polyhedral in shape, possessed a large spherical nucleus with heterochromatin. Leydig cells are typical steroid synthesizing cells with a rich network of smooth endoplasmic reticulum with tubular form cisternae, which represented the most remarkable, characteristic of these cells. The rough endoplasmic reticulum is formed of short cisternae with their associated ribosomes. The cytoplasm contains numerous mitochondria of various shapes and sizes which are frequently associated with the lipid droplets. The inner structure of the mitochondria consists of quite numerous tubular cristae, the mitochondrial matrix is moderately dense (Fig. 34). The testes of mice treated with repeated doses of reldan for 10 days (group M6) revealed marked ultrastructural changes in the seminiferous epithelium and its boundary tissues. The boundary tissue of the seminiferous tubule appeared thickened and folded. The myoid cells are swollen and their cytoplasm contained small vesicles, dense granules and vacuoles (Fig. 35). The electron micrographs (PL.6 & 7, Figs. 35 – 40) of

testicular tissue (group M6) revealed degenerated spermatogonia, spermatids are partially surrounded by Sertoli cell cytoplasm containing many vacuoles. The spermatozoa revealed high vacuolation of some late spermatids. Leydig cell exhibited ruptured contours with excentrically nuclei and large member of lysosomes, mitochondria and few lipid droplets were also seen. Moreover, degenerated, vacuolated, exfoliated and highly karyolytic nuclei with dense patches of nucleoli were clearly obvious in figure (40).

Many authors proved that reldan insecticide caused ultrastructural changes in both Sertoli and germinal cells of the seminiferous tubules of both mice and pigeons (*El-Wessemy, 2000*). *Gravis and Weaker, (1977)* suggested that the progressive folding, thickening of the peritubular tissue of seminiferous tubules may be a result of the progressive decrease in diameter of the seminiferous tubules. Also, the fibrous network and myoid cells have elastic properties which result in shrinkage and thickening of the peritubular tissue to accommodate the decrease in tubular diameter. On the other hand, *El-Sayyad et al. (1995)* stated that the alterations of testicular elements may be attributed to dysfunction of pituitary and testicular associated hormones as well as to the direct effects of the parent compound or its metabolites on testicular elements.

In conclusion, our results showed that such insecticides and soil fertilizers have marked hazardous impacts on the body organs, both structurally and functionally, so creating a serious threat to the life of man and his domestic animals. So, it is recommended that the use of these materials should be prohibited and substituted by safer agents in this concern.

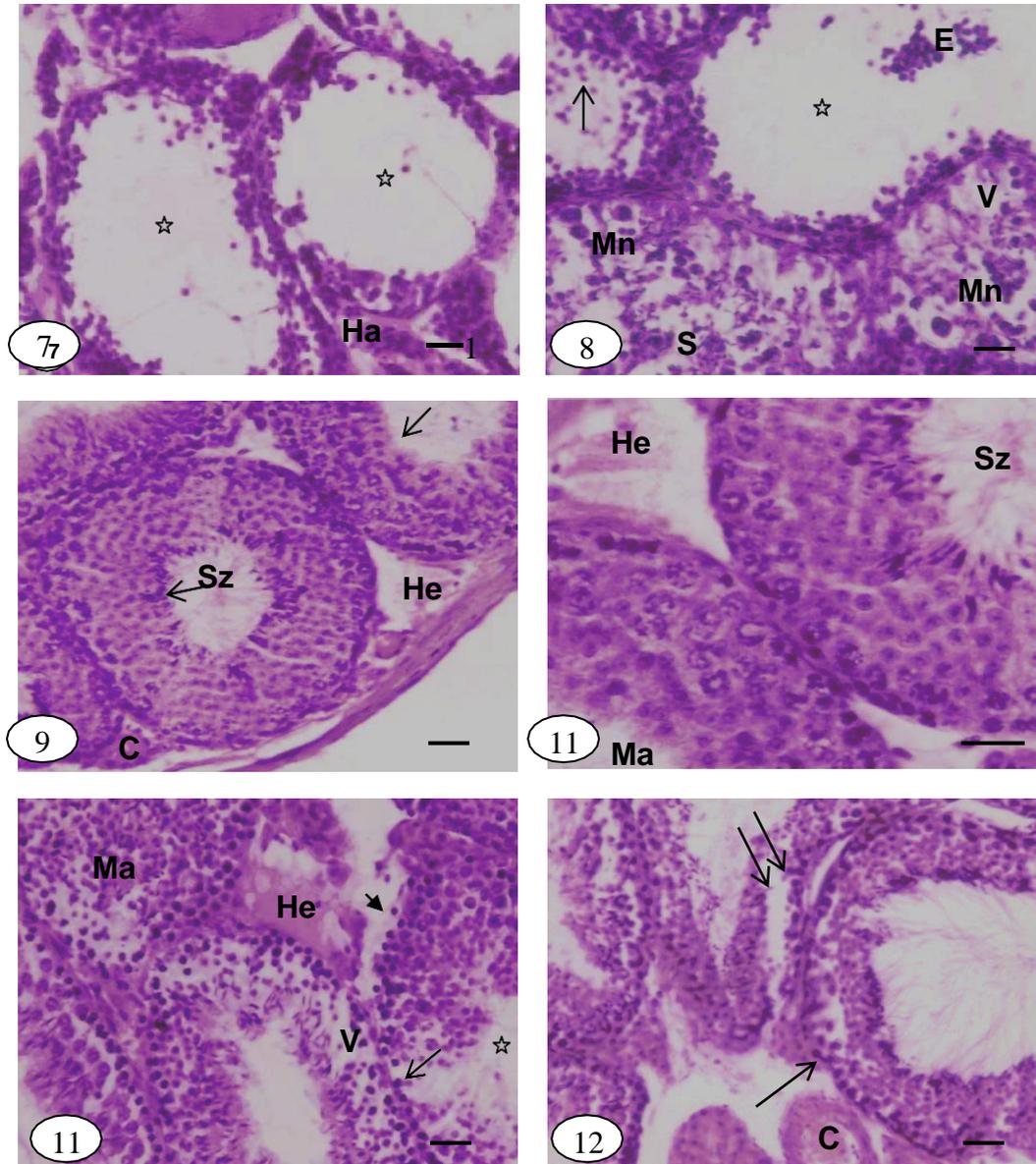
Plate (1)



Figs. (1-3): Sections of testes of control mice showing tunica albuginea (Ta), seminiferous tubules (St), interstitial tissue (arrow), spermatogonia (Sg), primary spermatocyte (Ps), spermatozoa (Sz), Sertoli cells (Sc) and leydig cells (Lc). Bars = 50 μ m & 20 μ m (Hx & E).

Figs. (4-6): Sections of testes of reldan treated-mice group M2 showing disruption of the basal lamina (arrow), hypoplasia * & ***, multinucleated cells (Mn) exfoliated (E) germ cells, maturation arrest (Ma), congestion (C) of blood vessels and vacuoles (V). Bars = 50 μ m (Hx & E).

Plate (2)

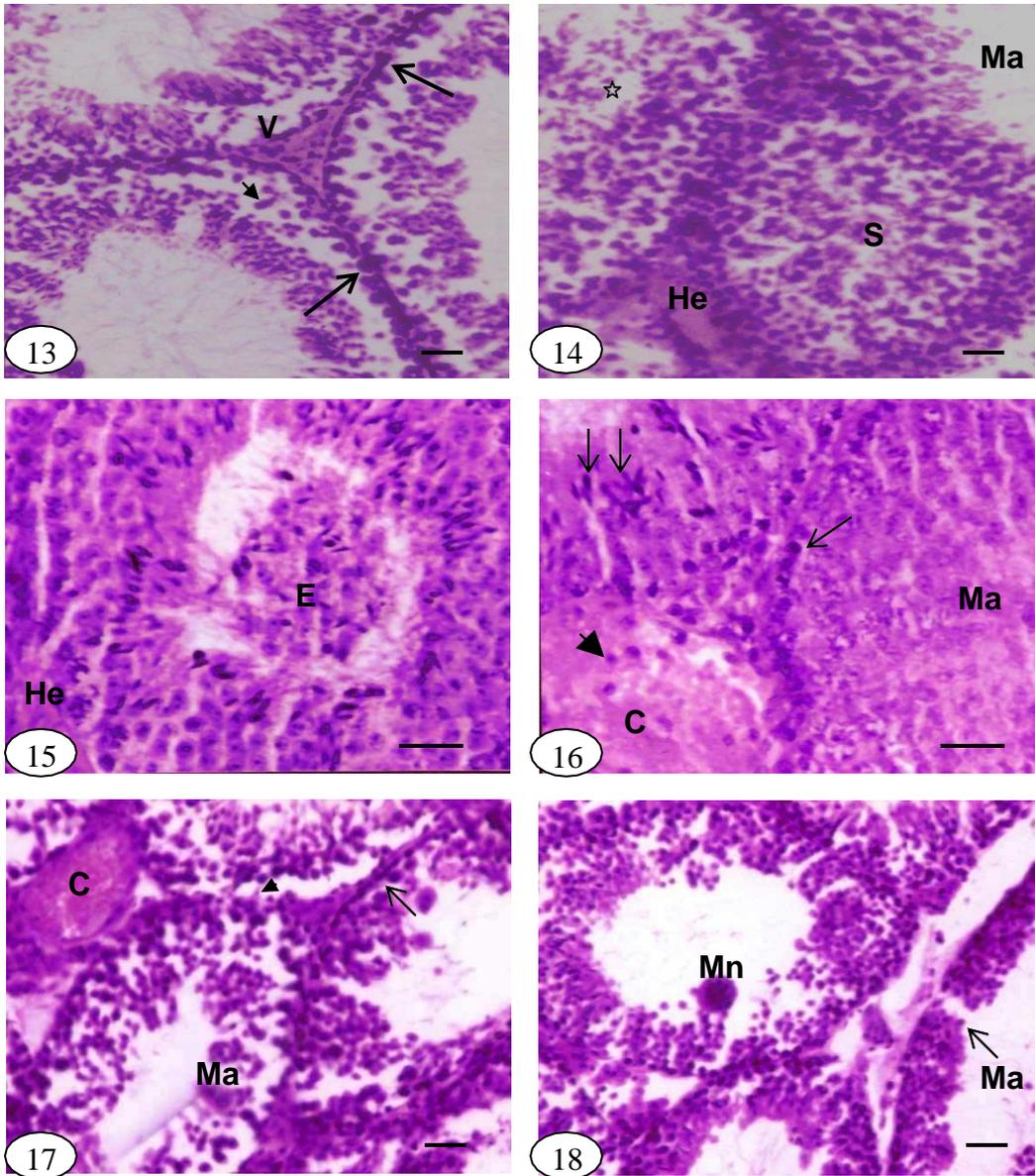


Figs. (7 & 8): Sections of testes of reldan treated-mice of group M3 showing hypoplasia of germinal cells hemorrhage (He), exfoliated (E) germ cells, multinucleated cells (Mn), vacuoles (V) and sloughing (S). Bars = 50 μ m & 20 μ m (Hx & E).

Figs. (9 & 10): Sections of testes of mice treated with a single oral dose of reldan (M4) showing spermatozoa (Sz) in the lumen of some tubules. Bars = 50 μ m & 20 μ m (Hx & E).

Figs. (11 & 12): Sections of testes of reldan treated-mice group M5 showing hypospermatogenesis (star), pyknotic unclai (arrow) and irregular contours of seminiferous tubules (double arrow). Bars = 50 μ m (Hx & E).

Plate (3)

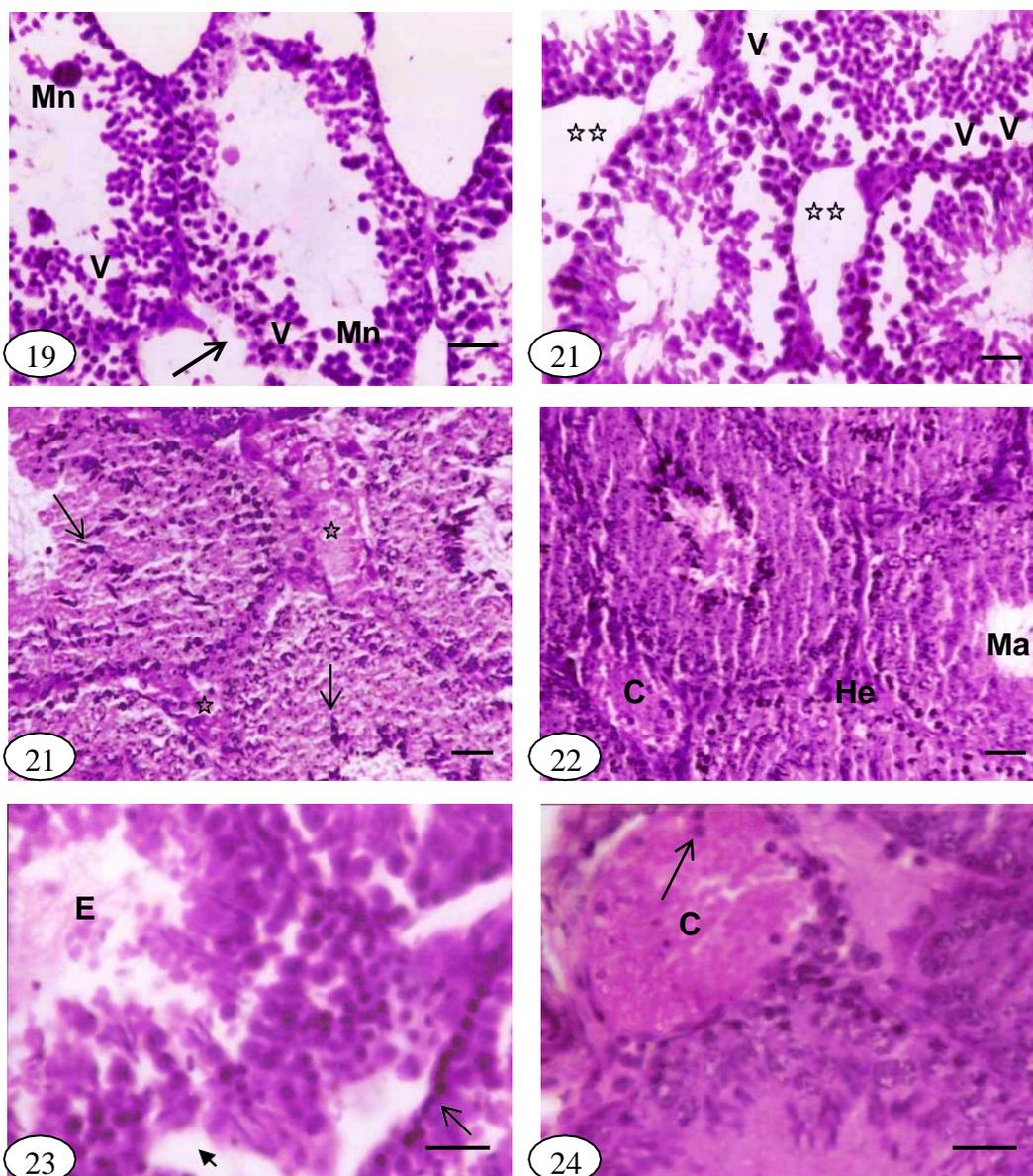


Figs. (13 & 14): Sections of testes of reldan treated-mice of group M6 showing the spermatocytes and spermatids separated from spermatogonial cells (arrow head), nuclear pyknosis (arrow), sloughing (S) and vacuoles (V). Bars = 50 μ m (Hx & E).

Figs. (15 & 16): Sections of testes of reldan treated-mice of group M7 showing slight degeneration of spermatogenesis, exfoliation (E), marked congestion (C) with inflammatory cells (arrow head). Bars = 50 μ m.

Figs. (17 & 18): Sections of testes of mice treated with ammonium nitrate (group M8) showing disruption of basal lamina (arrow head) and pyknosis (arrow). Bars = 50 μ m (Hx & E).

Plate (4)

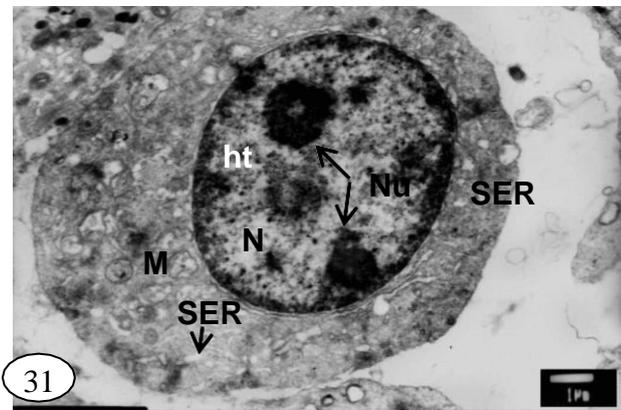
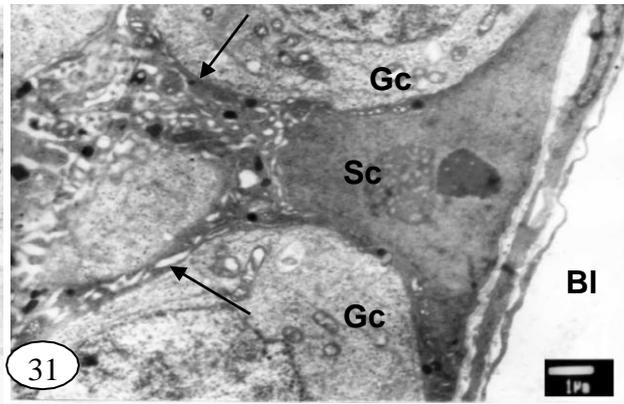
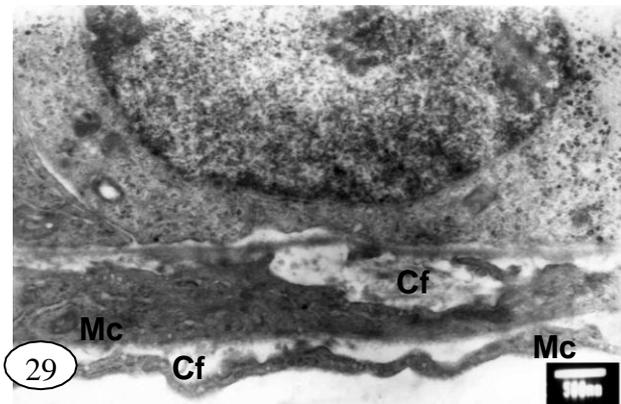
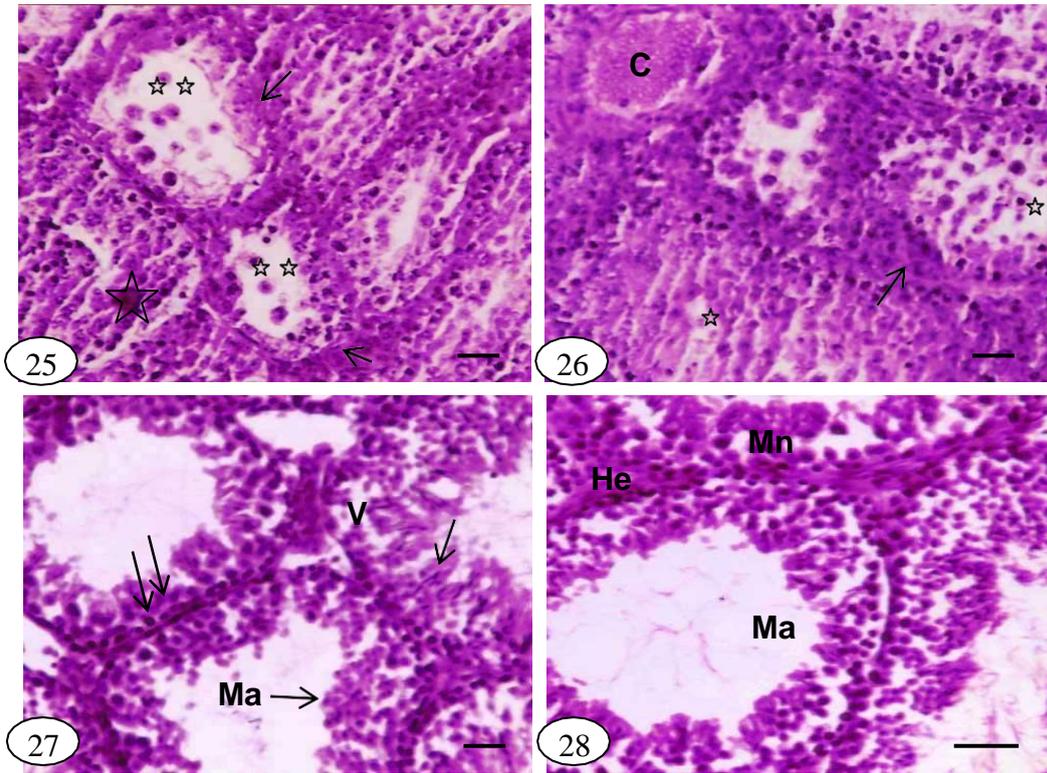


Figs. (19 & 20): Sections of testes of mice treated with ammonium nitrate of group M9 showing disruption of basal lamina (arrow), multinucleated giant cells (Mn), degenerated Leydig cells (***) and vacuoles (V). Bars = 50µm (Hx & E).

Figs. (21 & 22): Sections of testes of mice treated with ammonium nitrate (group M10) showing dilated blood vessels engorged with blood (star) and necrosis in spermatogonia (arrow). Bars = 50 µm.

Figs. (23 & 24): Sections of testes mice treated with ammonium nitrate (group M11) showing exfoliated cells (E), necrosis in spermatogonia (arrow) and congestion (C) of blood vessels. Bars = 50µm.

Plate (5)

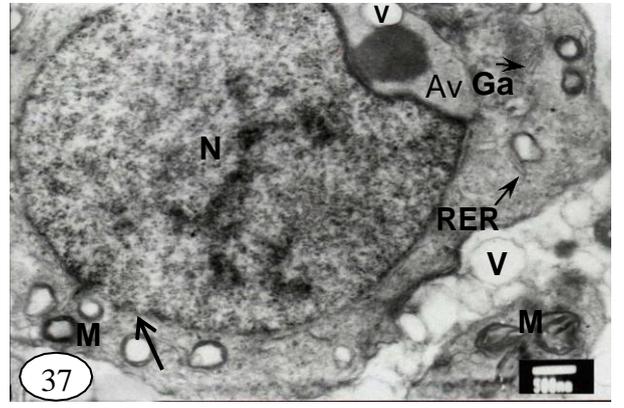
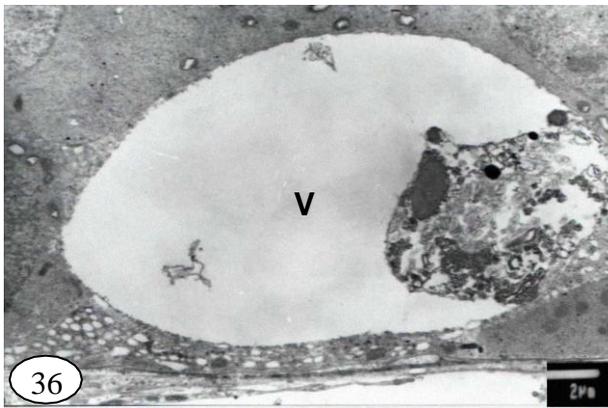
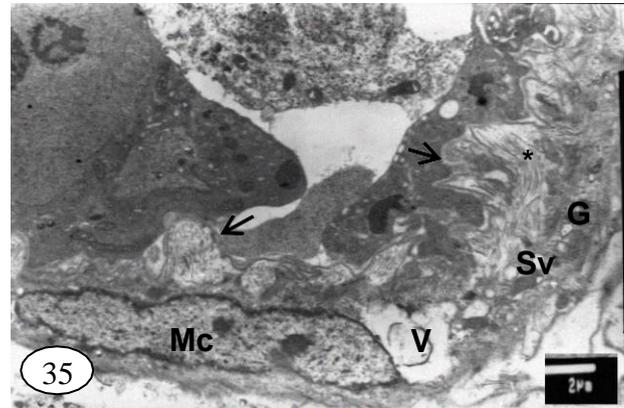
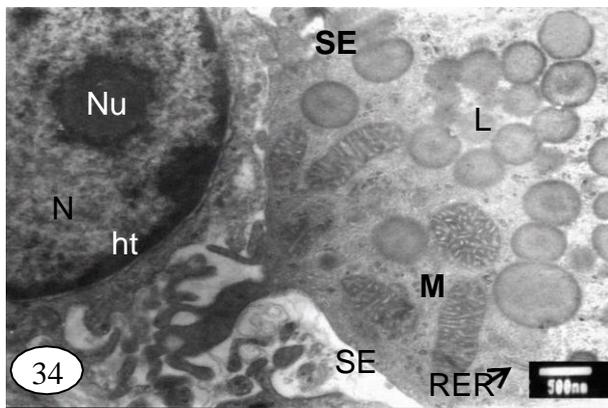
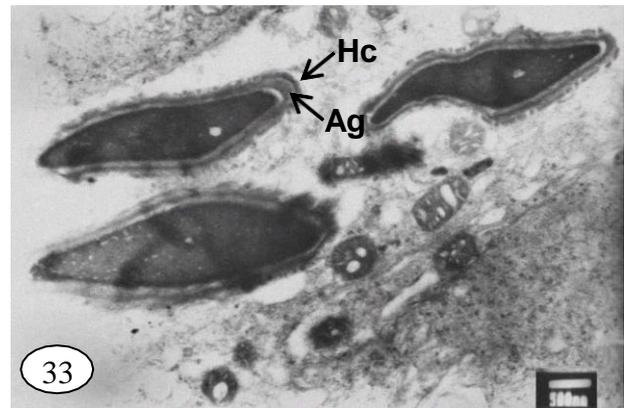
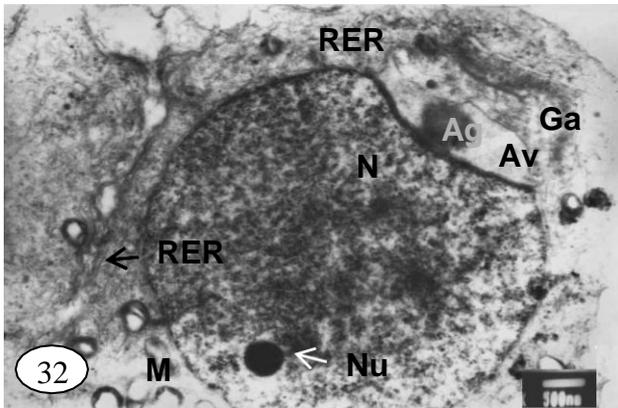


Figs. (25 & 26): Sections of testes of mice treated with ammonium nitrate of group M12.

Figs. (27 & 28): Sections of testes of mice treated with ammonium nitrate (group M13). Bars = 50 μ m.

Figs. (29 - 31): Electron micrograph (E.M.) of testes of control mice showing collagenous fibers (Cf) myoid cells (Mc), Sertoli cell (Sc) with lateral processes (arrows) surrounded the germ cells (Gc), also, spherical nucleus (N) contains heterochromatin (ht) of B-spermatogonium with mitochondria M, smooth endoplasmic reticulum (SER) in the cytoplasm. Bars = 50 μ m.

Plate (6)



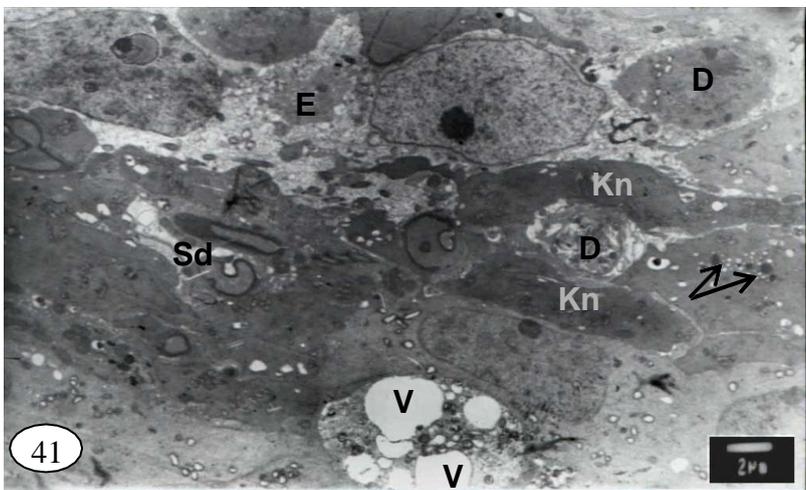
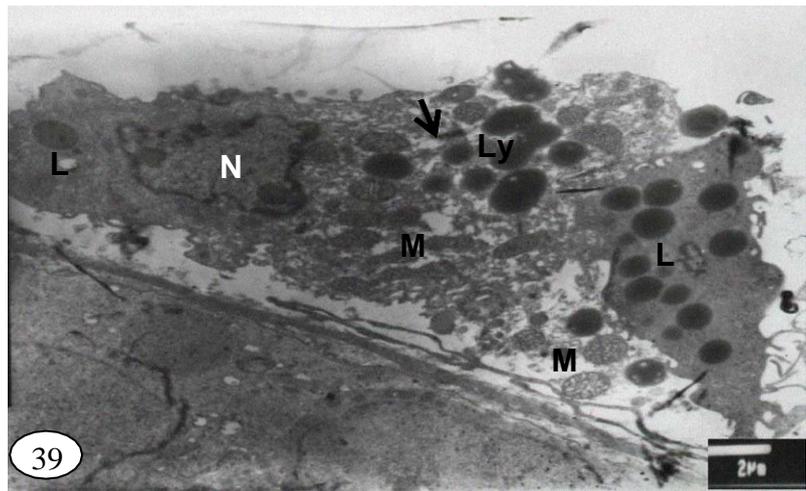
Figs. (32 - 34): E.M.showing the cap phase of spermatid contains nucleus (N) with heterochromatin (ht), mitochondria (M) and rough endoplasmic (RER) (Fig. 32). Also, the acrosomic phase of spermatids showing the head cap (Hc), acrosomal granules (Ag) (Fig. 33). Leydig cell of control mouse showing nucleus (N) nucleoli (Nu), its cytoplasm contains mitochondria (M) and lipid droplets (L) (Fig. 34)

Figs. (35 - 37): E.M.of testis of mice treated with a daily oral dose of reldan (1.2mg/kg.b.wt.) for 10 days (group M6), showing thickened (*) and folded (arrow) boundary tissue of seminiferous tubule. Also, the myoid cell (MC), small vesicles (Sv), dense granules (G) and vacuoles (V). Fig. (36) showing degenerated spermatogonial cell leaving a large empty space (V). Fig. (37) showing spermatid-surrounded by Sertoli cell cytoplasm containing vacuoles (V). Vacuoles in acrosomal vesicle (AV), well developed mitochondria (M) and Golgi apparatus (Ga). The nucleus (N) contains fewer nuclear materials with the disappearance of the outer nuclear membrane (arrow).

Plate (7)



Figs. (38 - 40): E.M. of testes of mice treated with a daily oral dose of reldan (1.2 mg/kg.b.wt.) for 10 days showing late spermatozoa with increased vacuolation (V) of some late spermatids. Cross and longitudinal sections of sperm tails surrounded by the cytoplasm of Sertoli cells. Fig. (39) showing Leydig cell with nuclei (N), lysosomes (Ly), mitochondria (M) and lipid droplets (L) were seen- Fig. (40) showing spermatogenic cells with degenerated (D), vacuolated (V) and exfoliated (E) sperms. Also highly karyolytic nuclei (Kn) with dense patches of nucleoli (arrows).



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الدوليدان ونترات الألمونيوم أحدثا تغيرات نسجية مرضية وتكبيرية دقيقة في خصية الفئران الصغيرة

محمود أحمد البندهاوي⁷، ناهد حسين رياض²، شادية على رضوان³، منى العقاد⁴، عزيزة الوسيدي⁵

7، 2، 4 قسم علم الحيوون - كلية العلوم - جامعة عين شمس

3، 5 قسم العلوم البيولوجية والجيولوجيا - كلية الشريعة - جامعة عين شمس

يهدف هذا البحث دراسة التغيرات الانسجائية في الخصية والتكبير الحقيقى الناتج عن انخفاض إفرازات
الخصية عن المعدل الطبيعي بالفئران المصابة بمرض السكري من النوع الثاني (DM2) في تصديق ارتباطها بالبنية
تكون جرتا عن مفرقة أ مكرتة من صرف ارتباط اح صرف يبرغ هلي ينف اح عضوى (بلفان) 72 م/ قيدي من
زن اجلبوان (أ) صرف ارتباط اح صرف يبرغ هلي ينف اح عضوى (بلفان) 72 م/ قيدي من زن اجلبوان (أ) صرف
19 أ 5 أيام من اح باح بل بوا اي اللعامل غ لفة 5 أيام بعف 24 ل اغ من آخرت جرتا اجلبوا اي
اللعامل غ لفة 19 أيام بعف 24 ل اغ أ 5 أيام من آخرت جرتا غ.
تهدف الى اخفام اجلبوان أ دداي الموم نادناي ا نض يبرغ مرتك برف ح ا a
انف ا م ا a ا ا ا ا ا ا ا ا ا ا ا a ا ا ا ا ا ا a ا ا ا a
جرف اجا ويغ ق ح ك ط ا لدا ا ا ا ا ا ا ا ا ا ا ا ا ا a ا ا ا ا ا ا ا ا ا ا ا a ا ا ا a ا ا a
اح يو ا ف مرتدلغ مع برف .
أيضاً أ ك ا ا ا ا ا ا ا ا ا ا ا ا ا ا ا ا a ا ا ا ا ا a ا ا ا a ا ا a ا ا a ا a ا a ا a ا a ا a ا a
يا ل ا ا ا ا ا ا ا ا ا ا ا ا ا a ا ا a ا a ا a ا a ا a ا a ا a ا a ا a ا a ا a ا a ا a ا a ا a ا a ا a
خ ا ا لرتنوا خ ا ا برفج) اح ا ا ا ا ا a .