The possible protective effect of vitamin E on adult albino rat's testes exposed to electromagnetic field emitted from a conventional cellular phone

Mahmoud Ibrahim EL-Naggar, Alaa EL-Din Sayed EL-Sagheer, Ashraf El- Sayed Ebaid.

Anatomy and Embryology department, Faculty of Medicine, Al- Azhar University

Abstract

Aim of the work: i to discuss the possible protective effect of vitamin E as a model of powerful antioxidant on the testes of adult albino rates exposed for electromagnetic field (EMF) emitted from cellular phone. This study also detected and described signs of morphological and behavior changes that can appear on the rats. **Materials and Methods:** The present study was carried out on 120 healthy adult albino rats. The rats were divided into 3 main groups. Group I (control group): consisted of 40 rats. These rats were kept in the animal house away from any source of EMF. Group II (radiation group): consisted of 40 rats that were exposed to EMF emitted from mobile phone for 60 days. Group III (radiation and vitamin group): consisted of 40 rats, those were exposed to EMF emitted from mobile phone for 60 days and simultaneously they received vitamin E, orally. **Results:** Electromagnetic field exposed rats showed testicular alterations, which were ameliorated by using vitamin E. **Keywords**: Mobile phones, electromagnetic field, vitamin E, testis.

INTRODUCTION

When the biological systems are exposed to an external magnetic field with a very large strength relative to the bio-magnetic field of the cells, a disturbance in their metabolic function is expected and may lead to death of the cells or increase their cell division ⁽¹⁾. Exposure of mice to 900-1800 MHz microwaves affected the histological structure of testis particularly Leydig cells and showed an apoptosis inducing effect on the spermatogenic cells ⁽²⁾. Free radical formation and their interaction with biological system is a matter of major concern. There is evidence of free radical generation after exposure to microwave radiation field ⁽³⁾. ROS generated by mobile phones exposure if not scavenged may lead to widespread lipid, protein and DNA damage ⁽⁴⁾. Electronic household items and cell phones are reported to decrease fertility potential in men by decreasing sperm count, motility, viability, and inducing pathological changes in sperm and testes morphology ⁽⁵⁾. Spermatogenesis is a complex process takes place in the testis, which may be exposed to various microwave frequencies, which are currently in use ⁽⁶⁾. Among various factors of infertility, oxidative stress has become the factor of interest as a potential cause of male infertility ⁽⁷⁾. The effect of EMF on human health varies widely depending on the frequency and intensity of the fields⁽⁸⁾.

There are numerous studies showing that EMF exposure of male rat/mice affects testicular structure, spermatogenesis, sperm motility, Leydig cell reduction, increased apoptosis of germ cells and in general subfertility and or infertility⁽⁹⁾.

Vitamin E is one of the major antioxidants; it plays an important role in reducing oxidative stress ⁽¹⁰⁾. Vitamin E suppresses lipid peroxidation in testicular microsomes and mitochondria. It reverses the detrimental effects of oxidative stress on the testicular function ⁽¹¹⁾. Vitamin E is associated with normal function of the male reproductive system. Supplementation with vitamin E has also been shown to increase sperm concentration, improve sperm motility, enhances sperm and semen quality ^(1,13).

MATERIALS AND METHODS Material:

Experimental animals.

The present study was carried out on 120 healthy adult male albino rats weighing from 200 to 300 grams each, obtained from the Egyptian Holding Company for Biological Products and Vaccines (Cairo, Egypt). All rats were kept in clean properly ventilated cages under similar conditions and had free access to laboratory food and water throughout the experiment. The rats were divide into 3 main groups:

Group I (control group): consisted of 40 rats. These rats were kept in the animal house away from any source of EMF.

Group II (radiation group): consisted of 40 rats that exposed to EMF emitted from mobile phone for 60 days.

Group III (radiation and vitamin group): consisted of 40 rats, those were exposed to EMF emitted from mobile phone for 60 days and simultaneously they received vitamin E 1.35 mg/ kg, body weight, 3 times/week, orally (two hours before exposure to EMF) $^{(14,15)}$.

1) Vitamin E: Vitamin E was obtained as a concentrate of DI-alpha-tocopheryl acetate (soft gelatin capsules 400 mg) from Pharco Pharmaceutical Company, Cairo Egypt. The concentrate was dissolved in distilled water. Dissolution of fluid aspirated from gelatinous capsules of vitamin was done in pharmacology department, AI-Azhar faculty of pharmacy (16).

Methods:

- 1) Electromagnetic wave exposure: Rats of groups II and III were exposed to EMF emitted from mobile phone (TECNO T470). Group II of rats were exposed only to EMF emitted from mobile phone for 60 days. Rats of group III were exposed to EMF for 60 days, and simultaneously they received vitamin E orally (two hours before exposure to EMFs). During EMF exposure, the cages were arranged in a circular manner and the mobile phone was placed in the center. The mobile phone was ringed for 120 min/day for the whole period of the experiment. This was done by ringing 24 times/30 min (using auto redial application for android mobiles from Google play). The time of ring is 50 seconds with 10 seconds interval between each two successive rings (14,15).
- 2) Measurements and Behavior: Mean body weight of each animal was estimated at beginning of the study and every 2 weeks, while mean of the body gain was identified at the end of Sections of the control rat testes showed complete spermatogenesis was established

complete spermatogenesis was established, spermatogonia were seen close to the basement membrane with, primary spermatocytes were the largest cells, the spermatids appeared smaller than primary spermatocytes and lying near the lumen. The lumen enclosed a large sperms and some of these sperms were attached experiment. The mean of testicular weight was recorded at the end of experiment. The changes in the behavior of animals also were documented.

- **3)** Collection of Blood Sample: Blood samples were collected through the medial canthus into EDTA bottles for hormonal assay ⁽¹⁷⁾.
- **4) Hormonal Assay:** Plasma samples were assayed for testosterone using the enzyme-linked immunosorbent assay (ELISA) technique.
- **5) Semen Collection:** The testes were removed along with the epididymis. The caudal epididymis were separated from the testes, blotted with filter papers and lacerated to collect the semen⁽¹⁷⁾.
- **6)** Semen Analysis: Sperm morphology and count.
- 7) Histopathological examination: Testis tissues were submitted for histological and morphologic examinations using conventional methods and were stained with hematoxylin eosin and hematoxylin iron.
- **8)** Statistical analysis: The obtained data were analyzed to determine the significance of differences among means. Then data were expressed as mean ± standard deviation. Values will consider significant at p<0.05.

RESULTS

Microscopic Appearance Light microscopic Group I (control group):

to the apex of Sertoli cells. Testicular tissue of control animals showed average sized tubules, complete spermatogenesis, average interstitium and average tunica albuginea. Seminiferous tubules of the testes possessed epithelia containing the Sertoli cells and the germ cells of various stages, covering the complete process of spermatogenesis (**Figs. 1- 4**).



Figure (1): Light microscopic photomicrograph of section in the testis of group I (control group) showing average sized tubules (T) with average germinal lining (blue arrow), complete spermatogenesis (black arrows) and average interstitium (red arrow) (H&E X 200).



Figure (2): Light microscopic photomicrograph of section in the testis of group I (control group) showing seminiferous tubules lined with series of spermatogenic cells, spermatogonia (green arrow), primary spermatocyte (black arrow), and spermatids (red arrow). Sertoli cells (yellow arrow), are seen with attached many spermatozoa (blue arrows). Tubules are surrounded by average basement membrane enclosing myoid cells (violet arrow). The interstitial spaces in between the tubules contains interstitial cell of Leydig (white arrow). (H&E X 400).



Figure (3): Light microscopic photomicrograph of section in the testis of group I (control group) showing seminiferous tubule with average BM (violet arrow), Sertoli cells (yellow arrow) appeared tall cells with

pale nucleus with prominent nucleolus, spermatogonia (green arrow) with their dark nuclei with prominent nucleolus; primary spermatocyte (black arrow) is the largest cell with dark condensed nucleus. The spermatids (white arrow) appeared smaller than primary spermatocytes and lying near the lumen. The lumen enclosed a large sperms (blue arrows) and some of these sperms were attached to the apex of Sertoli cells. Average interstitium showing Leydig cells (red arrow) having vesicular nucleus with prominent nucleolus. (H&E X 1000).



Figure (4): Light microscopic photomicrograph of section in the testis of group I (control group) showing seminiferous tubules with average BM (violet arrow), Sertoli cells (blue arrow), spermatogonia (green arrow), primary spermatocyte (black arrow) and many spermatozoa (red arrows) (Iron hematoxylin X 400).

Group II (radiation group):

Testicular tissue of radiation exposed animals showed disorganization, degeneration, decreased diameters and atrophy in some seminiferous tubules. Markedly distorted tubules showed irregular configurations, thin germinal lining, detached BM and intra-tubular desquamated cells. There were widely spaced distorted irregular seminiferous tubules with marked interstitial edema, congested blood vessel and marked sub-capsular edema. There was increase in interstitial spaces that contained few interstitial cells of Leydig with marked edema. Degeneration of the epithelium was common in the majority of the seminiferous tubules as there was marked reduction of spermatogenesis and partial loss of basement membrane. Marked reduction of germinal lining composed mainly of spermatogonia and primary spermatocytes with few sperm. The effects were more severe in spermatid differentiation. The lumen contained germ cell debris and fewer spermatozoa (Figs. 5-8).



Figure (5): Light microscopic photomicrograph of section in the testis of group II (radiation group) showing widely spaced distorted seminiferous tubules (black arrows) (H&E X 200).



Figure (6): Light microscopic photomicrograph of section in the testis of group II (radiation group) showing average tubules with thin germinal lining (blue arrow), the seminiferous tubules showing vacuolated spermatogonia (yellow arrow), vacuolated primary spermatocytes (green arrow) and spermatids (red arrow). Marked interstitial edema with vacuolation (violet arrow) showing Leydig cells (black arrows) had scanty cytoplasm with deeply stained or normal vesicular nuclei (H&E X 400).

Group III (radiation and vitamin group):

Examination of the testis in this group revealed somewhat normal appearance of most seminiferous tubules and Leydig cells. In spite of normal appearance of some seminiferous tubules, some showed delamination of spermatogenic layers from the basement



Figure Light microscopic (7): photomicrograph of section in the testis of group II (radiation group) showing the basement membrane was damaged and irregular (blue arrow), distorted tubule with marked reduction of germinal lining composed mainly of spermatogonia type A dark (green arrow), type A dark (olive green), type B (pink arrow), few primary spermatocytes (black arrow) and spermatids (violet arrow) with very few sperms (red arrow). There is marked degeneration of spermatogenic cells leading to increased intra-cellular spaces (white arrow) with intra-luminal desquamated epithelial cells (yellow arrow) (Iron hematoxylin X 1000).



Figure (7): Light microscopic photomicrograph of section in the testis of group (radiation Π group) showing seminiferous tubules with thin germinal lining with detached BM (violet arrow), increased intra-cellular spaces (yellow arrow), spermatogonia (green arrow) and primary spermatocyte (black arrow) with reduction of spermatozoa (red arrows) (Iron hematoxylin X 400).

membranes. The architecture of seminiferous tubules was nearly preserved, average sized tubules, complete spermatogenesis, average tunica albuginea. The basement membrane of tubules was slightly corrugated with subcapsular edema and markedly congested blood vessel. In many tubules, there were normal spermatogonia, normal primary spermatocytes and Sertoli cells with few attached mature sperms. Some distorted tubule showed mild reduction of spermatogenesis, with thin germinal lining composed mainly of spermatogonia and primary spermatocyte with reduction of spermatozoa. Interstitial edema in some tubules showed Leydig cells having scanty cytoplasm and normal vesicular nuclei (**Figs. 9- 11**).



Figure (8): Light microscopic photomicrograph of section in the testis of group III (radiation and vitamin group) showing tunica albuginea (blue arrow), marked sub-capsular (black arrow) and interstitial (red arrow) edema (H&E X 200).



Figure (9): Light microscopic photomicrograph of section in the testis of group III (radiation and vitamin group) showing normal basement membrane of tubules (red arrow), average tubules with and germinal lining complete average (blue arrow). spermatogenesis Marked interstitial edema showing Leydig cells (black arrow) had scanty cytoplasm with normal vesicular nuclei and prominent nucleolus. (H&E X 400).

Electron microscopic

Electron micrographs of control animals revealed that the seminiferous tubule was surrounded by a basement membrane and lined



Light Figure (10): microscopic photomicrograph of section in the testis of group III (radiation and vitamin group) showing seminiferous tubules with slightly BM (violet arrow). disrupted Normal spermatogonia type A dark (green arrow), type A pale (white arrow), type B (olive green arrow). Normal primary spermatocyte (black arrow), spermatids (red arrow), with Sertoli cells (yellow arrow) with vesicular nucleus, and few attached mature spermatozoa (blue arrow) (H&E X 1000).



Figure (11): Light microscopic photomicrograph of section in the testis of group III (radiation and vitamin group) showing seminiferous tubules with thin lining composed germinal mainly of spermatogonia (green arrow) and primary spermatocyte (black arrow) with reduction of spermatozoa (red arrows) (Iron hematoxylin X 400).

Group I (control group):

by spermatogenic epithelium. The epithelium showed the usual sequence of spermatogonia, primary spermatocytes and spermatids. Sertoli cells and germ cells with cellular characteristics were typical of those seen in active spermatogenesis. The germ cells were in various developmental stages and the cells laving on basement membrane showed flat myoid cell (Fig. 12). Sertoli cells showed distinct nucleus and cytoplasmic characteristics consistent with an active secretory state. It rests on the basement membrane of the tubule, extending towards the lumen of the tubule, filling the narrow spaces between the cells of the spermatogenic series. (Figs. 12 and 13).



Figure (12): Transmission electron micrograph of a section in the testis of control group revealing Sertoli cell with pyramidal shaped large euchromatic nucleus (N) with prominent nucleolus (Nu) and enclosed with nuclear envelope that exhibiting a deep indentation (DI). The cytoplasm contains mitochondria. Spermatogonium (SG) type B with rounded cytoplasm nucleus (n). the contains mitochondria (m), and the cells lying on basement membrane (BM). (x5000)



Figure (13): Transmission electron micrograph of a section in the testis of control group revealing basal part of Sertoli cell (SC) contains mitochondria (M). Basal part of spermatogonium (SG) type B with nucleus (n) and enclosed with nuclear membrane (Nm).

The spermatogonia rest upon the basement membrane of the tubules. The primary spermatocytes were rounded in configurations with prominent large rounded nuclei (Fig. 14). The elongated spermatids were manifested by the formation of the acrosome in developing spermatid. (Fig. 15). Parts of sperms appeared also. (Fig. 16).

The cell membrane (CM) appears intact, these cells lying on basement membrane (BM) that showing myoid cell (MC) (x10000)



Figure (14): Transmission electron micrograph of a section in the testis of control group revealing primary spermatocytes with fine chromatin nucleus (N), prominent nuclear membrane (Nm) appears normal and their cytoplasm contains rounded mitochondria. (x 6000)



100000

Figure (15): An electron micrograph of a section in the testis of control group showing early changes in spermatid into sperm (spermiogenesis), elongated spermatid having large oval nucleus (N) possessing euchromatin (Eu), the nuclear membrane thickened from one side forming acrosomal cap (Ac) over the anterior hemisphere of the nucleus. The cytoplasm contains microtubules (MT), mitochondria (M) on other side. (x 10000)



Figure (16): An electron micrograph of a section in the testis of control group showing change of spermatid into sperm (spermiogenesis), the longitudinal sections for the head and neck (n) of sperm, the head is formed of condensed nucleus (N) covered by acrosomal cap (AC). With ectoplasmic specialization (ES).Transverse section (TS) of midpiece of tail appears. (x 10000)

Group II (radiation group):

In this group, some seminiferous tubules showed degenerative changes in the developing spermatogenic epithelium, irregular basement membrane of seminiferous tubule. Vacuolization was evident in the cytoplasm of the Sertoli cells, spermatocytes and spermatids. Sertoli cell nucleus irregular in shape, decrease size and indented. Intra-cytoplasmic in vacuoles with increased intercellular spaces between cells. Spermatogonia rests on thick membrane lost irregular their normal architecture (Figs. 17 and 18). Primary spermatocytes, which are rounded in shape, showed abnormal clumping of chromatin and vacuolated deformed mitochondria. All cells cytoplasmic vacuolations revealed and rarification with increased intercellular spaces (Fig. 19). Rounded spermatids were distorted and degenerated. The cytoplasm showed vacuolated mitochondria deformed and lamellar body (Fig. 20). Few and incompletely developed sperms were observed in the tubular lumen.



Figure (17): An electron micrograph of a section in the testis of 2^{nd} (radiation) group showing irregular basement membrane (BM) of seminiferous tubule. Sertoli cell nucleus (n) irregular in shape, decrease in size and indented. Its cytoplasm showing mitochondria and intracytoplasmic vacuoles (V) with increased intercellular spaces (ICS) between cells. Spermatogonium showing irregular nuclear and cellular outlines. (x 5000)



THH Hag - 10000m

Figure (18): An electron micrograph of a section in the testis of 2^{nd} (radiation) group showing thick irregular basement membrane (BM) with myoid cell (MC). Spermatogonium possessing irregular indented nucleus (N), the cytoplasm contains mitochondria (M) and showing irregular cell membrane. A part of Sertoli cell appears (SC) showing vacuoles (V) with intracellular spaces (ICS). (x 10000)



2 BLOWMEN THE MING - DOBOK

Figure (19): An electron micrograph of a section in the testis of 2nd (radiation) group showing thick irregular basement membrane (BM) of seminiferous tubule. Spermatogonia (SG) possessing rounded nucleus-containing euchromatin marginated (Eu) and heterochromatin (Ht). Sertoli cell shows part of its pyramidal shaped nucleus that enveloped by intact nuclear membrane with slight irregularity (I), its cytoplasm showing intracytoplasmic vacuoles (V) with increased intercellular spaces (ICS) between cells. Primary spermatocytes (PS) which are rounded in shape having large rounded nucleus (ns) showing abnormal clumping of chromatin (CH) with ill-defined nuclear membrane (Nm) and irregular cell

The majority of the seminiferous tubules showed regular basement membrane, nearly normal histological appearance of spermatogenic epithelium. The Sertoli cells and germ cells were normal. The Sertoli cells, spermatogonia, spermatocytes and spermatids, round as well as elongated appeared normal. The basement membrane of the tubule is covered by fibrous connective tissue, with an innermost layer containing myoid cell. Spermatogonia possessing rounded to oval nuclei, containing nucleoli. Intranuclear and



Figure (21): An electron micrograph of a section in the testis of 3^{rd} (radiation + vitamin) group showing slightly irregular basement

membrane (CM). Vacuolated mitochondria (M) are seen clearly in the cytoplasm. All cells reveal cytoplasmic vacuolation and rarification (degenerative changes). (x 5000)



2 8107-080 TEM Hag = 100408

Figure (20): An electron micrograph of a section in the testis of 2^{nd} (radiation) group showing rounded spermatid with nucleus (N) irregular nuclear membrane (Nm) outline of spermatid having aproacrosomal granule (AG) and acrosomal cap (AC) with absence of microtubules and mitochondria. The cytoplasm contains vacuoles, cristae distortion of mitochondria (M) and lamellar bodies (LB). Longitudinal section of head appears showing condensed nucleus (n) and acrosomal cap (ac) covering anterior part of the head. (x 10000)

Group III (radiation and vitamin group)

intracytoplasmic vacuolations may be present with intracellular spaces (Figs. 21 and 22). Primary spermatocytes having large rounded nucleus, surrounded by nuclear membrane. The cytoplasm contains oval mitochondria (Fig. 23). Regarding spermatids, some were intact and others were less developed, normal developing spermatids possessing large rounded nucleus, surrounded by nuclear membrane that showing a proacrosomal granule not detached from nuclear membrane. (Fig. 24).

membrane (BM); spermatogonia (SG) possessing rounded to oval nuclei (n), and the cytoplasm containing mitochondria (m). Sertoli cell (SC) which is pyramidal in shape. Their nuclei are typically ovoid or triangular (N), euchromatic (Eu), and have a prominent nucleolus (Nu). Numerous mitochondria (M) seen clearly in the cytoplasm. Its nuclear membrane (Nm) and cell membrane (Cm) appears intact. (x5000)



Figure (22): An electron micrograph of a section in the testis of 3^{rd} (radiation + vitamin) group is showing spermatogonia (SG) possessing rounded nucleus (N), and its nuclear membrane (Nm) looks disturbed. There are intracellular spaces (ICS), the basement membrane of the tubule is covered by fibrous connective tissue, with an innermost layer containing flattened, smooth muscle like myoid cells (MC) possessing flat nucleus (n). (x10000)



Figure (23): An electron micrograph of a section in the testis of 3^{rd} (radiation + vitamin) group showing primary spermatocyte having large rounded nucleus (N) containing nucleolus (Nu), and surrounded by nuclear membrane (Nm). Cytoplasmic organelles, however, are poorly defined. The plasma membranes of the spermatocytes are distinct, contain oval mitochondria (M) and show closer association with the Sertoli cell. (x6000)



Figure (24): An electron micrograph of a section in the testis of 3^{rd} (radiation + vitamin) group showing elongated spermatid having large oval nucleus (N). The nuclear membrane is thickened from one side forming acrosomal cap (Ac) over the anterior hemisphere of the cytoplasm contains nucleus. The oval mitochondria (M) and rough endoplasmic reticulum (RER). Degenerated spermatid (DS) is seen showing atrophy in size, pyknotic nucleus having condensed chromatin materials and the surrounding cytoplasm containing deformed vacuolated mitochondria (V). (x10000)

DISCUSSION

The current study showed that exposure of male rats to 950 MHz EMF 3 hours per day, for two months induced disorganization, degeneration and atrophy in some seminiferous tubules with dilated interstitial spaces and highly reduced Leydig cells. Delaminated spermatogenic cells from the basement membrane, ruptured basement membranes of some seminiferous tubules, decreased spermatogenic layers and spermatogenic cells with highly reduced sperms in the lumen of the seminiferous tubules were also realized. EMF of 950 MHz radiation selected in the present experiment because it is a frequency of the Global System Mobile (GSM) signal modulation used for all mobile communication (18).

Mobile phone radiation results in increased oxidative stress, with subsequent sperm membrane lipid and DNA damage. These abnormalities seem to be directly related to the duration of mobile phone use $^{(19)}$.

In the present study, the testicular damage in radiation-exposed rats was indicative for harmful effect of EMFs. Seminiferous tubules have various degrees of degeneration of spermatogonia with the presence of numerous abnormal changes in all spermatogenic cells. These findings are in agreement with that were reported by *Al-Damegh* ⁽¹¹⁾; she observed depletion of the spermatogenic cells and decrease in the epithelial height of seminiferous tubules in here work. Her results are coinciding with our results.

On the other hand, the increased epithelial height of rats protected with vitamin E compared to radiation exposed group proved vitamin E protective effect against radiation possible oxidative effect. The findings of our study are commonly linked to testicular oxidative stress induced by radiation. Al-**Damegh** ⁽¹¹⁾; found that electromagnetic radiation from a conventional cellular telephone cause marked elevation of malondialdehyde (MDA) level in testicular sample homogenate.

In the present study, morphologically abnormal sperms were revealed in rat testis exposed to radiation. *Venkatesh et al.* ⁽²⁰⁾, documented ROS induced abnormal sperm morphology. Radiation induced ROS production could lead sperm membrane lipioperoxidation, mitochondria damage and consequently, increased sperm deformity index ⁽²¹⁾.

Cytoskeletal inhibition by radiation leads to decreased cholesterol uptake by Leydig cells and resultant lowered testosterone synthesis. Additionally, radiation induced cytoskeletal dysfunction could down regulate synthesis and/or transport of LH receptors to Leydig cells plasma membrane ⁽¹¹⁾. The reduction in serum testosterone is documented to be accompanied by the histopathological changes that are represented by production of high numbers of apoptotic cells, spermatogenic cells disintegration and decreased Leydig cell viability. This could explain the findings of the current study.

Antioxidants are the main defense factors against oxidative stress induced by free radicals ⁽²²⁾. Vitamin E is the key antioxidant component of spermatozoa and membrane protectants against ROS ⁽²³⁾, and could be considered as a good prophylactic agent against radiation induced toxicity. In the current work, testis of radiation exposed rats, protected by vitamin E showed that the architecture of most of seminiferous tubules was highly preserved with enhanced spermatogenesis. The lumen of the tubule was full of sperms. Interstitial cells of Leydig appeared having normal vesicular nuclei and abundant cytoplasm. There was also proper differentiation of spermatids with acrosomal cap and acrosomal vesicle. Most of sperms were morphologically normal. Our findings are supported by what is stated by *Amr and Noaman* ⁽¹⁵⁾, *who* also assured vitamin E protective role against hazardous effect of radiation.

Thus, the increased radiation induced free radicals generation in testes might have been neutralized by vitamin $E^{(11)}$, as vitamin E is chain breaking antioxidant. This may be enforced by other antioxidants chemicals, such as vitamin C, that are important for regenerating antioxidant ability of alphatocopherol⁽²⁴⁾. In vitro experiments performed by *Ross et al.*⁽²⁵⁾. It is proved that vitamin E protects spermatozoa from oxidative damage. It enhances sperm motility and performance. This is also, confirmed later by *Liu et al.*⁽²⁶⁾ and *Ourique et al.*⁽²⁷⁾.

Results of the present experiment showed that single administration of vitamin E to rats for 2 months before exposure to 950 MHz of EMF resulted in remarkable regenerative features as most of the seminiferous tubules and Leydig cells retained their normal histological appearance in spite of the presence of numerous empty spaces in between the spermatogenic cells with normal arrangement of the spermatogenic layers and cells with reappearance of mature sperms.

The current observations come in agreement with the previous reports of *Jacobs* et al $^{(28)}$ and Oral et al $^{(29)}$ who reported that antioxidants could protect living cells from radiation damage by ameliorating the deleterious effect of free radicals. Also supplementation with antioxidant (vitamin E) scavenging reactive oxygen species and increasing antioxidant enzyme activities and prevent 950 MHz EMF-induced oxidative damage of liver and heart $^{(30)}$.

Lastly, the current study could conclude that radiation causes degenerative changes of the testes of albino rats and arrest of spermatogenesis. The radiation induced histological and ultra-structural changes of the testes. Moreover, it could be concluded, that vitamin E has a protective role against radiation-induced damage in testicles by its antioxidant and anti-apoptotic effects. Vitamin E protective effect is reflected on testicular histology and ultra-structure.

REFERENCES

- 1. Horiuchi S, Ishizaki Y, Okuno K, Ano T and Shoda M. (2002): Change in broth culture is associated with significant suppression of E. coli death under high magnetic field. Bioelectrochemistry, 57: 139-144.
- 2. *Nassar SA* (2009): Do microwaves of mobile phone affect the testicular tissue structure? (Histopathological and ultrastructural study). Egypt. J. Hosp. Med., 37: 685- 699.
- 3. D'Angelo C, Costantini E, Kamal MA and Reale M (2015): Experimental model for ELF- EMF exposure: Concern for human health. Saudi Journal of Biological Sciences, 22: 75– 84.
- 4. Jajte J, Grzegorczyk J, Zmyslony M and Rajkowska E (2002): Effect of 7 mT static magnetic field and iron ions on rat lymphocytes: apoptosis, necrosis and free radical processes. Bioelectrochemistry, 57:107-111.
- Erogul O, Aydur E, Komesli G, Irkilata HC, Irmak MK and Peker AF. (2006): Effects of electromagnetic radiation from a cellular phone on human sperm motility: an in vitro study. Arch. Med. Res., 37(7): 840-843.
- 6. Behari J and Kesari KK (2006): Effects of microwave radiations on reproductive system of male rats. Embryo Talk, 1 (1): 81-85.
- Kumar S, Kesari KK and Behari J (2010): Evaluation of genotoxic effect in male wistar rats following microwave exposure. Ind J. Exp. Biol., 48: 586-592.
- 8. *Genuis SJ (2007):* Fielding a current idea: Exploring the public health impact of electromagnetic radiation. Public Health, 18.
- 9. *Khaki AA, Tubbs RS, Farahani RM, Zarrintan S and Nag TC (2006):* The effects of an electromagnetic field on the boundary tissue of the seminiferous tubules of the rat: a light and transmission electron microscope study. Folia Morphol., 65: 105-110.

- Aydogan F, Atılgan HI, Koca G, Sadiç M, Korkmaz M, Tuncal S and Samim E (2014): An evaluation of the radioprotective effect of vitamin E on the salivary glands of radioactive iodine in rats. Turkish. J. Ear, Nose and Throat, 24 (1): 21-29.
- 11. Al-Damegh MA (2012): Rat testicular impairment induced by electromagnetic radiation from a conventional cellular telephone and the protective effects of the antioxidants vitamins C and E. Clinics, 67: 785-792.
- 12. Uzunhisarcikli M, Kalender Y, Dirican K, Kalender S, Ogutcu A and Buyukko MF (2007): Acute, subacute and subchronic administration of methyl parathion induced testicular damage in male rats and protective role of vitamins C and E. Pesticide Biochemistry and Physiology, 87: 115-122..
- **13.** *Mohamed AK (2014):* The possible rescue effect of vitamin E or silymarin on lung tissues of male albino rats exposed to electro-magnetic field. Egypt. J. Hosp. Med., 57: 470-481.
- 14. Jaarin K, Gapor MT, Nafeeza MI and Fauzee AM (2002): Effect of various doses of palm vitamin E and tocopherol on aspirin-induced gastric lesions in rats. Int. J. Exp. Pathol., 83: 295-30.
- **15.** Ahmet A, Nevres HA, Tolga A and Selçuk Ç (2011): The effects of electromagnetic field exposure at short and long term of 900 MHz frequency emitted from mobile phones on rat bone tissue, 38 (4): 452-457.
- **16.** *Amr MA and Noaman AE (2015):* The possible protective effect of vitamin E and/or silymarin on rat testes exposed to 950MHz electromagnetic field, Journal of Bioscience and Applied Research, 1:97-111.
- 17. Oyedeji KO, Bolarinwa AF and Adigun AK (2013): Effect of Aspirin on Reproductive Functions in Male Albino Rats. Journal of Pharmacy and Biological Sciences (JPBS), 4: 49-54.
- Khalil A, Al-Adhammi M, Al-Shara B, Gagga M, Rawshdeh A and Al-Shamli A(2012): Histological and ultrastructural analyses of male mice exposed to mobile phone radiation. J. Toxicol. Rev., 1: 1-6.

- **19.** La vignera S, Condorelli RA, Vicari E, D'agata R and Calogero AE (2012): Effects of the exposure to mobile phones on male reproduction: A review of the literature. J. Andrology, 33(3):350-356.
- 20. Venkatesh S, Singh G, Gupta N, Kumar R, Deecaraman M and Dada R (2009): Correlation of sperm morphology and oxidative stress in infertile men. Iran J Reprod Med., 7: 29–34.
- 21. Das J, Ghosh J, Manna P, Sinha M and Sil PC (2009): Taurine protects rat testes against NaAsO (2)- induced oxidative stress and apoptosis via mitochondrial dependent and independent pathways. Toxicol Lett.,187: 201–10.
- 22. Agarwal A, Prabakaran SA and Said TM (2005): Prevention of oxidative stress injury to sperm. J Androl., 26:654–60.
- **23.** *Yousef MI, Abdallah GA and Kamel K* (2003): Effect of ascorbic acid and vitamin E supplementation on semen quality and biochemical parameters of male rabbits. Anim Reprod Sci., 76: 99–111.
- 24. *Traber MG (2012):* Vitamin E, Modern Nutrition in Health and Disease, 9th edition. Baltimore, MD: Williams & Wilkins.
- 25. Ross C, Morriss A, Khairy M, Khalaf Y, Braude P, Coomarasamy A and et

al. (2010): A systematic review of the effect of oral antioxidants on male infertility. Reprod Biomed., 20:711–23.

26. Liu Q, Zhou Y, Duan R, Wei H, Jiang S and Peng J (2016): Lower dietary n-6: n-3 ratio and high-dose vitamin E supplementation improve sperm morphology and oxidative stress in boars.

www.publish.csiro.au/rd/rd15424

- 27. Ourique GM, Saccol EM, Pês TS, Glanzner WG, Schiefelbein SH, WoehlVM and et al. (2016): Protective effect of vitamin E on sperm motility andoxidative stress in valproic acid treated rats. Food Chem Toxicol. 95:159–67.
- 28. Jacobs BC, Dennehy G, Ramirez J, Sapp J and Lawrence VA (2002): Milk thistle for the treatment of liver diseases: a systematic review and metaanalysis. Am. J. Med., 113: 506-515.
- 29. Oral B, Guney M, Ozguner F, Karahan N, Mungan ., Comlekci S and Cesur G (2006): Endometrial apoptosis induced by a 900-MHz mobile phone: preventive effects of vitamins E and C. Adv. Ther., 23 (6):957-973.
- 30. Ibrahim NK and Gharib OA (2010): The Protective effect of antioxidants on oxidative stress in rats exposed to the 950 MHz electromagnetic field .J. Rad. Res. Appl. Sci., 11: 45- 51.