# Biochemical and hematological effects of electromagnetic field on male rats.

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## ABSTRACT

The present study was performed to investigate the effect of electromagnetic field (EMF) on some biochemical and hematological parameters in rats. Thirty white male albino rats 12-16 weeks old divided into two groups (15 each): the first group used as control and the second was the experimental group kept in specific test cell (this cell was designed specially for use in that experiment). Rats were exposed to the electromagnetic field for about 18 hr every day only for 3 months. Blood samples were collected from all rats after 1, 2, and 3 months from exposure to EMF in the morning from medial canthus of eyes. Samples were collected on tubes containing EDTA and divided into two parts. One part used for separation of plasma and RBCs. Plasma was used for determination of creatinine phosphokinase (CK), lactate dehydrogenase (LDH), monoamino oxidase (MAO) and acetyl cholinesterase (Ach E), as well as cortisone hormone, malondialdhyde (MDA), nitric oxide (NO) and haptoglobin were also determined. While RBCs used in preparation of hemolysate which was used for estimation of antioxidant enzymes activities. The other part still as whole blood for hematological examination. Experimental group showed alteration in serum enzymatic activities as compared to control. Moreover, increased in the serum level of MDA and NO was detected which reflected changes in the activity of biological system.

**Key words:** EMF,Ach E , MAO, CK, LDH, MDA, and cortisone.

## INTRODUCTION

With the ever increasing use of electronic devices, extremely low frequency electromagnetic fields (EMF) have become a fact of modern life. These fields are given off by all electronic devices including high energy sources like power lines ,microwaves, home wiring airport, and transformers **(Van Deventer *et al.*, 2005)**.Moreover, the biological effects of extremely low frequency electromagnetic fields have been concerned that the children living in homes with an excess of electrical wiring configuration suggestive of high current flow had a higher incidence of cancer **(Chen et al., 2000)**, depression **(Lyer et al., 2003**) birth and reproduction anomalies **(Blaasaas et al., 2003**), brain tumor, leukemia, miscarriage, chronic fatigue, headache, cataracts, heart problems, stress, nausea, chest pain, forgetfulness

and other health problems **(Mercola., 2009)**.In addition to decreased membrane enzymes activities as alkaline phosphatase, acetylecholinestrase and phosphoglycerate kinase **(Morelli et al., 2005),** also exposure to magnetic field may lead to physiological changes particularly in the field of mood disorders where the 5-hydroxyl tryptamine (5-HT) system is strongly involved. The precise mechanism underlying these effects is not known but the consensus is that extremely low frequency field (ELF) interact with biological systems through electric fields, either applied or induced by time varying magnetic fields **(Liburdy., 1995)**. In recent years using of mobile phones exposed us to low –intensity electromagnetic radiation which play a role of that reactive oxygen species in EMF induced oxidative damage in tissue which is evident by the increase in MDA and NO level **(Ilhan et al., 2004)**.Therfore, the present study was applied to investigate the effect of EMF used with frequency exposure in useful devices on human biological system.

## MATERIALS AND METHODS

Thirty female albino rats, 12-16 weeks old, weighted 150-200g were used in the present experiment. Rats were obtained from animal house, Faculty of Veterinary Medicine, Benha University. All animals were kept under constant environment and nutritional condition throughout the experimental period. Water was supplied ad-libitum.

**Test cell:** consisted of two plates of copper. One is fixed and the other is movable. The distance between the two plates is 50 cm, one was earthed and the other was connected to supply. Cell dimension was 30 and 20 cm. two sides of the cell were made of wood and other parallel sides were made of fiber, between the two copper plates there was barrier of insulating material fixed to protect rats to touch energized copper plate. Cell was energized from the supply through autotransformer. A resistance of 14 ohm was connected as a load. The current flows to the test cell through upper plate. Timer was used to control the time of energized the test cell **(Waleed, 2008)**. Rats were divided into two groups (15 each).

**Group (1):** acted as control and kept in the same environmental condition like the other group.

**Group (2):** the exposed group which was kept inside the test cell and was exposed to the electrical and magnetic field for about 18hr every day which adjacent by a timer and disconnected the supply for about 6hr daily. This process was repeated for 3 months. Autotransformer output was fixed at 220v, while a variable resistance was fixed at 14ohm to feed cell by a current of 16A. That means current field=1100V and magnetic field=2.66A/m.

Blood samples were collected from all rats after 1, 2 and 3 months after exposure to EMF in the morning from medial canthus of eyes on EDTA and divided into two parts. One part used for separation of plasma and RBCs by centrifugation.

Plasma directed for estimation of CK **(Kachmar and Moss 1976)**, LDH **(Buhl and Jackson 1978),** AChE **(Denblown 1983)**, MAO **(Mc Eween, 1969),** cortisol **(Wilson, 1977),** MDA **(Esterbaur, *et al.* 1982)**, NO **(Montogomery & Dymock, 1961)** and haptaglobin **(Kurosky, 1980)**. Erythrocytes were washed 3 times by normal saline solution and processed for estimation of antioxidant enzymes activities such as glutathione reductase (GR-ase) **(Bergmayer, 1983)**, glutathione-S- transferase (GST) **(Habig and Jakaby, 1974)**, glutathione peroxidase (GSH-Px) **(Chiu et al., 1976),** catalase **(Sinha, 1972)** and superoxide dismutase (SOD) **(Misra and Fridovich, 1972),** reduced glutathione (GSH) concentration **(Beultar, *et al.* 1983)** the other part still as whole blood for determination of hematological parameters were determined by using hemocytometer method for RBCs count, Wintrobe macrohematocrite method for PCV and Drabkin method for Hb determination according to **Drabkin, (1932).**

**Statistical analysis:** The obtained data were statistically analyzed and the significant difference between groups evaluated by t-test as explained by **Snedecor and Cochran, (1982).**

## RESULTS

**Table (1):** revealed a non significant increase in plasma activities of CK and LDH at 1 and 2 month which become significant increase after 3 months as exposure to EMF compared to control group. Rats exposed to EMF showed also a significant decrease in plasma AChE activity allover the experimental period, while MAO revealed non significant changes compared to control.

A non significant decrease was recorded in plasma cortisone level after one month that decrease became significant after 2 and 3 months from exposure to EMF, plasma MDA levels revealed a highly significant increase at one month, that increase became significant after 2 and 3 months. Also, NO level showed significant increase after 1, 2 and 3 month of EMF exposure when compared to control group.

Plasma haptoglobin level showed a non significant increase after 1, 2 and 3 months from exposure to EMF when compared with control group.

**Table (2):** revealed that rats exposed to EMF showed anon significant changes in the erythrocytes GSH concentration as well as, GR-ase, GST and catalase activities after 1, 2, and 3 months as compared to control group. While SOD and GSH-Px were significantly decreased after 3 months of expoure to EMF.

**Table (3):** showed that, no significant differences were observed in the hematological parameters (RBCs, Hb, PCV, MCV, MCH, MCHC and WBCs) between the control group and the exposed group after one, two and three months of EMF exposure.

## DISCUSSION

High electric current, computers, mobile phones and their bases station are an important source of ultrahigh frequency electromagnetic field and their utilization is increasing allover the world. Epidemiological studies have suggested that low energy may have biological effects such as changes in oxidative metabolism after exposure **(Ferreira *et al.*, 2006).** The detected significant increase of both CK and LDH levels after 3 months of exposure to EMF are agreed with these of **Zhang *et al*., (2000)** who reported that, electromagnetic shield applied to volunteers showed a highly significant effect on serum level of creatinine phosphokinase. Moreover, **(Olson and David, 1984)** reported an elevation in CK, LDH and GOT after exposure to radiofrequency. This may be due to that magnetic fields interact with moving charges in cells and change their velocities, (**Goodman and Blank, 2002)**. However, Cell membranes have been identified as a primary site of interaction with the low frequency fields **(Adey, 1988).** Therefore, the alterations in these charges and molecules consider the first step in the production of biological effects as magnetic field interact with moving charges and change enzymatic activity **(Vizcaino, 2003**). In addition, EMF may regulate the rate and the amount of product of biochemical reaction possibly through free radical mechanism including direct influence on enzyme action **(Till *et al.*, 1998)** in contrast, **(Horakowa *et al.*, 2005)** stated that electric field has no affect on plasma creatinine phosphokinase activity in rats.

Regarding the significant decrease of plasma acetylcholinesterase enzyme our results are similar to that of **(Hillert et al., 2001)** who reported that, people exposed to EMF revealed marked reduction in cholinesterase activity. Moreover**, (Morelli et al., 2005)** recorded that EMF caused a significantly decrease in the enzymatic activity of acetylcholinestrase however, the phenomena apparently reversible.

Exposure of rats to EMF (80Ka/M) resulted in an increased of cholinesterase activity in cardiac structure, beside neurocytes. While a continuous application to EMF for long time resulted in a progressive decrease of cholinesterase activity **(Abramov and Merkulova 1980)** This may be attributed that EMF has significant correlation to difficulties in concentration of enzyme in muscle, as AchE inhibited by free radicals such as OH and H2O2 (**Tsakiris et al., 2000)**

However, MAO is an enzyme involved in brain catabolism of monoamine neurotransmittor with oxidative deamination results in production of hydrogen peroxide derived from MAO activity represent special source of oxidative stress in the brain **(Soto-otero et al., 2001).** The recorded non significant change of plasma MAO activity was come inaccordance with **(Dolgacheva et al., 2000)** who noted that, the effect of ultra low power pulse- modulated electromagnetic radiation on the activity of MAO enzyme involved in the oxidative deamination of monoamino. It was established that, the increase of activity MAO in hypothalamus reached the maximal

meaning at modulation frequency of 6Hz while modulation frequency of 20Hz the activity of enzyme was decreased. **(Kami et al., 1985)**

The observed significant decrease in plasma level of cortisone of EMF exposed rats was similar to the results of **(Bonhomne et al., 1998)** who showed that, EMF lead to decrease in cortisol level which may be attributed to an increase in ACTH and cortisol values at high doses and long exposure, and later feed back control with symptoms like acute adrenal insufficiency due to an inhibition of hypothalamic pituitary adrenocortical function.

Concerning the significant elevation of plasma MDA and NO levels. Our results are agreed with **(Guler et al., 2008)** who reported that, low frequency electric field has potential harmful effects on living organism by enhancing the radical production which indicated by a significant elevation in the level of antioxidant product as MDA and NO. However, Malondialdhyde is the break down of the major chain leading to oxidation of polyunsaturated fatty acid, thus it serve as a reliable marker of oxidative stress mediated per oxidation **(Serel et al., 2004).** the higher rate of oxidative metabolic activity, and higher concentration of readily oxidizable membrane polyunsaturated fatty acid **(Meral et al., 2007)**. EMF prolongs the life of free radicals and can act as a promoter or copromotor of cancer **(Coskun et al., 2006).**

The significant increase of NO may be declared that, magnetic field may be act through sequential steps, initial imbalance in iron homeostasis that increase free iron levels and leads to formation of hydroxyl radicals via the Fenton reaction, follwed by lipid peroxidation processes and calcium leakage from internal storage then trigger activity of nitric oxide synthesis and the release of NO which responsible for damage to DNA and other macromolecular districts **(Lai and Singh, 2004)**. thus changes in NO, MDA levels indicate increased ROS production occurring during exposure period that, may reflect the pathological process of EMF exposure. **(Fehmi et al., 2005**).

The recorded non significant increase of plasma haptoglobin level was agreed with **(Madsen et al., 2002)** who stated that, as haptoglobin is acute phase protein in nature any stress factor as EMF may increase the levels of plasma haptoglobin. But the real way for this mechanism and effect of EMF on haptaglobin need more researches.

The observed changes in the antioxidant defense system are similar to the data obtained by **(Turkzer et al., 2008)** who recorded non statistically significant difference in antioxidant defense mechanisms between rats exposed to electromagnetic field and control. Also, **(Moustafa et al., 2001)** showed that, the activity of SOD and GSH-Px in human erythrocytes revealed significant decrease while catalase didn't significantly decrease. These results indicated that, acute exposure to EMF may modulate the oxidative stress of free radicals by enhancing

lipid peroxidation and reducing the activation of SOD and GSH-Px which are free radical scavenger. As magnetic field penetrates the cells and can alter cell membrane potential and the concentration of ions **(Canseven et al., 2005)**. These alterations may affect radical processes within the cell as free radicals formation induces change in enzyme activity, gene expression and alteration of membrane structure **(Khadir et al., 1999)**. Moreover, exposure to EMF result in deterioration of RBCs function and metabolic activity, it was expected that, the increase of toxicity in specific organs was a result of the RBCs functional failure. Therefore, changes in antioxidants may be due to the deterioration in cellular membrane properties in the liver. In addition to increase toxicity in different organs **(Qui et al., 2004).** As well as, decreases of SOD activity results in accumulation of superoxide anion radicals in blood. **(Kula et al., 2000).**

On contrast to **(Eraslan et al., 2007)** reported that, chronically applied of EMF to mice doesn't cause oxidative damage which indicated by not change of SOD, GSH-Px and catalase.

No significant difference was noted between the control and the exposed group as regards the blood parameters and the total leukocytic count at any experimental period. These results agree with the results obtained by **Bonhomme-Faivre et al. (1998)** after 63 and 90 days of treatment. On the other hand, the obtained results are on contrast to the results obtained by **Sedehi Esfahani et al. (2007)** who recorded a significant increase in RBCs, PCV and Hb in some groups of the electromagnetic field exposed rats but that may be the effect of chronic exposure (one year) applied by these authors.

From all these results we noticed that, EMF has side effect on biological system of body and its metabolism. So we advised with non frequent exposure to EMF for trying to decrease free radical release, change in cell charge which results in change in enzymatic activities. This may need more details by applying EMF for long time for more demonstrations of their effect.

**Acknowledgement:** The authors are grateful to the stuff members of the Power Department, High institute of Technology, Benha University specially **Dr/ Waleed** for their help in the design of test cell used in the search.

**Table (1): Effect of EMF on plasma, CK, LDH, AchE, MAO activities (U/L), cortisone(ug/dl),MDA(nmol/ml), NO(umol/l) and haptaglobin(mg/dl) in male rats.**

| Duration | Parameter group | CK | LDH | AchE | MAO | cortis one | MDA | NO | Hapto globin |
|---|---|---|---|---|---|---|---|---|---|
| One month | Control group | 435.07± 18.8 | 1353.33 ± 20.29 | 449.55± 16.94 | 15.07± 1.68 | 5.23± 0.53 | 29.86± 1.36 | 57.37± 1.67 | 6.57± 0.2 |
| | Exposed group | 375.97± 17.27 | 1426.67 ± 20.29 | 242.39± 18.09* | 19 ± 2.3 | 3.46± 0.78 | 44.72± 0.48** | 80.77± 0.74* | 7.8 ± 0.15 |
| Second month | Control group | 420.01± 17.09 | 1775.33 ± 15.78 | 426.53± 14.11 | 20.72± 0.37 | 5.47± 0.32 | 27.02± 2.21 | 52.74± 0.27 | 6.87± 0.2 |

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|---|---|---|---|---|---|---|---|---|---|
| | **Exposed group** | 467.97± 10.53 | 1315.33 ± 23.93 | 261.74± 12.42* | 18.76± 1.68 | 3.14± 0.18* | 45.42± 0.92* | 79.77± 2.05** | 8.23± 0.29 |
| **Third month** | **Control group** | 330.33± 15.29 | 1323.33 ± 17.66 | 403.55± 14.69 | 26.37± 0.94 | 7.3 ± 0.46 | 26.17± 4.17 | 48.08± 1.12 | 7.47± 0.26 |
| | **Exposed group** | 512.63± 14.61* | 1551.0± 22.83* | 267.74± 21.8*1 | 18.52± 5.52 | 5.08± 0.14* | 46.22± 2.34* | 78.77± 3.36* | 9.2 ± 0.59 |

Results are presented as mean ±S.E.

* (P< 0.05) ** (P< 0.01) Significant from control

**Table (2): Effect of EMF on erythrocyte on GR-ase, GST, GSH-PX, Gatalase, SOD activities (u/g protein) and GSH concentration (umol/g protein) of male rats.**

| Duration | Parameter group | GR-ase | GST | GSH-Px | catalase | SOD | GSH |
|---|---|---|---|---|---|---|---|
| **One month** | Control group | 9.71± 0.46 | 4.18± 0.2 | 46.93± 2.25 | 20.17± 0.44 | 120.03± 5.28 | 9.29± 0.42 |
| | Exposed group | 9.27± 0.32 | 3.99± 0.14 | 40.49± 1.59 | 19.47± 0.09 | 110.22± 3.71 | 8.87± 0.32 |
| **Second month** | Control group | 11.24± 0.81 | 4.84± 0.35 | 44.29± 3.9 | 20.06± 0.22 | 127.28± 9.14 | 10.75± 0.77 |
| | Exposed group | 11.49± 1.43 | 5.08± 0.55 | 41.76± 3.34 | 19.52± 0.18 | 119.47± 3.42 | 11.28± 1.23 |
| **Third month** | Control group | 12.76± 1.54 | 5.49± 0.63 | 42.64± 3.44 | 20.39± 3.22 | 127.39± 3.22 | 12.21± 1.25 |
| | Exposed group | 13.02± 1.48 | 5.61± 0.64 | 25.96± 3.18* | 19.53± 0.88 | 106.03± 3.87* | 12.47± 1.42 |

Results are presented as mean ±S.E.

* (P< 0.05) ** (P< 0.01) Significant from control

**Table (3): Effect of EMF on hematological parameters of male rats.**

| Duration | Group | RBCs ($10^6$/ul) | Hb (gm/dl) | PCV (%) | MCV (fl) | MCH (pg) | MCHC (%) | WBCs ($10^3$/ul) |
|---|---|---|---|---|---|---|---|---|
| **First month** | Control group | 3.57 ± 0.09 | 10.17 ± 0.30 | 31.50 ± 0.90 | 88.47 ± 0.38 | 28.55 ± 0.14 | 32.27 ± 0.03 | 4.47 ± 0.07 |
| | Exposed group | 3.69 ± 0.12 | 10.60 ± 0.46 | 32.80 ± 1.37 | 87.14 ± 0.95 | 28.71 ± 0.28 | 32.31 ± 0.04 | 3.93 ± 0.18 |
| **Second month** | Control group | 3.46 ± 0.09 | 10.13 ± 0.29 | 31.20 ± 1.04 | 90.05 ± 0.77 | 29.25 ± 0.11 | 32.02 ± 0.58 | 4.57 ± 0.26 |
| | Exposed group | 3.62. ± 0.07 | 10.35 ± 0.22 | 32.38 ± 0.70 | 89.36 ± 0.82 | 28.56 ± 0.13 | 31.97 ± 0.33 | 4.07 ± 0.24 |

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|---|---|---|---|---|---|---|---|---|
| **Third month** | Control group | 4.26 ± 0.19 | 12.3 ± 0.64 | 38.23 ± 1.63 | 89.77 ±1.01 | 28.86 ± 0.56 | 32.14 ± 0.34 | 3.87 ± 0.47 |
| | Exposed group | 3.83 ± 0.12 | 11.42 ± 0.55 | 35.25 ± 2.65 | 91.80 ± 4.82 | 29.76 ± 0.65 | 32.52 ± 1.02 | 5.17 ± 0.29 |

Results are presented as mean ±S.E.

SE= standared error.

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# التغيرات البيوكيميائية والفسيولوجية للموجات الكهرومغناطيسية علي ذكور الفئران البيضاء

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**الملخص العربى**

أصبح استخدام التكنولوجيا الحديثة إحدى المظاهر الأساسية المصاحبة للتطور العصرى داخل المنازل و خارجها كاستخدام الميكروويف و الكمبيوتر و التليفزيون والتليفون المحمول و جميع هذه الوسائل يعتمد تصميمها و عملها على الموجات الكهرومغناطيسية التى تخرج منها. ولما لهذه الموجات من أثار جانبية على الجسم البشرى. لذا كان الهدف من هذه الدراسة هو توضيح بعض الآثار الجانبية التى يتعرض لها الجسم البشرى داخل المجتمع من خلال تقييم الوظائف المختلفة للأعضاء. أجريت هذه الدراسة على عدد 30 من ذكور الفئران البيضاء قسمت الى مجموعتين رئيسيتين المجموعة الأولى: المجموعة الضابطة لم يتم تعرضها للموجات الكهرومغناطيسية ، المجموعة الثانية: (تحت الاختبار) والتى تم وضعها فى خلية الاختبار المخصصة حيث تتعرض فيها الفئران للموجات الكهرومغناطيسية لمدة 18 ساعة يوما وذلك لمدة ثلاث شهور متتالية. تم تجميع عينات الدم بعد شهر واثنين وثلاثة شهور متتالية من بداية التجربة علي مضاد تجلط EDITA وقد قسمت عينة الدم إلي جزئين الجزء الأول تم استخدامه لفصل البلازما وكرات الدم الحمراء ثم استخدام البلازما فى قياس كلا من إنزيم الكرياتين فوسفوكيناز ، اللاكتات ديهيدروجيناز ، المونوأمينوأومسيداز ، الكولين أستراز ، هرمون الكورتيزون ، مالون دايألدهيد ، أكسيد النيتريت ، الهبتاجلوبين. أما كرات الدم الحمراء تم تجهيزها لقياس الإنزيمات المضادة للأكسدة بها. و قد أوضحت النتائج: زيادة معنوية فى الشهر الثالث لإنزيمى الكرياتين فوسفوكيناز واللاكتات ديهيدروجيناز. أما أنزيم و المونوأمينوأومسيداز أحدث تغييرات غير معنوية فى حين ان أنزيم الكولين أستراز وهرمون الكورتيزون أظهرا نقص معنوى بالمقارنة بالمجموعة الضابطة.

كما سجلت النتائج زيادة معنوية في المالون داى الدهيد وأكسيد النيتريت خلال فترة التجربة. أما الهبتاجلوبين فقد أظهر تغيرات غير معنوية. أظهرت الإنزيمات المضادة للأكسدة فى كرات الدم الحمراء نقص معنوي فى نشاط إنزيم الجلوتاثيون بروكسيداز ، السوبر أوكسيد ديسميوتازو في حين أصبح هذا النقص غير معنوي مع الجلوتاثيون المختزل ، الجلوتاثيون ترنسفيراز ، الجلوتاثيون ريدكتاز ، اللاكتاز خلال فترة التجربة. و من هذه النتائج نستنتج أن للموجات الكهرومغناطيسية تحدث أثار جانبية على جسم الإنسان و لكى نتجنب حدوث هذه الآثار يجب أن لا نتعرض لفترات طويلة لإشعاع الأجهزة الحديثة حتى نتلافى الإصابة بالأمراض.