

Biochemical and biophysical variations in the characteristics of rat blood exposed to magnetic fields "in vivo study"

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Abstract: To study the effects of magnetic field (MF) of strength 50Hz-5mT on some biochemical properties of rat tissue. Forty eight of male albino rats were used, of average weight 170 ± 10 gm. The animals were housed in the same environmental conditions in plastic cages, and feed with balanced diet and tap water. The animals were divided into two groups as follows: Group A: Consists of 8 animals used as a sham radiation group and housed at normal environmental conditions of pressure and temperature. The temperature inside the lab varied between 22°C and 25°C during the experimental period. Lighting condition was day light and darkness during night. Group B: Consists of 40 animal was divided into five subgroups (8 animals for each group) namely B₁, B₂, B₃, B₄ and B₅, which were exposed to the magnetic field of 5 mT for a period of one week, two weeks, three weeks, and four weeks respectively. Group B₅ was exposed for a period of four weeks and used for performing the required analysis after 45 days (for delayed effect study). The results indicated that there is a significant decrease in the hematological Constituents of blood such as haemoglobin concentration Hb, hematocrit percentage and red blood cells count RBCs as compared to control group, but there is an improvement for the recovery group B₅. The viscosity of the blood was increased for animals of all groups as compared with control group A. The differences in viscosity demonstrate the effects of RBCs aggregation and deformability respectively. The result showed that the changes in liver enzymes and total protein from blood serum analysis showed that MF produced alteration in biochemical parameters of the liver transaminases SGOT and SGPT which have been widely utilized in mammalian toxicology as biomarkers of specific organ dysfunction. In general the increase in transaminases activity is usually associated with hepatocyte damage. While the kidney functions revealed a significant increase in the concentration of urea and creatinine compared with the control group (non-exposed). The increase of the concentration of urea and creatinine causes chronic complications in some organs of the body. It will be concluded that this study suggests that, in humans under investigation, the activities of liver enzymes GOT and GPT may increase by exposure to MF generated during magnetic resonance imaging or nuclear magnetic resonance procedures.

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Keywords: magnetic field, RBCs, SGOT, SGPT, urea, creatinine

1. Introduction

Electromagnetic fields (EMFs) are emitted from any source of energy, whether a simple heated metal pan, an electric wire, a flashlight, or the fusion or fission of two atomic nuclei. These waves are a combination of an electric field coupled with a magnetic field (Cember., 2008)¹.

Fields of different frequencies interact with matter and biological systems in different ways. The energy of ELF-EMFs is too low to cause ionization of molecules, i.e., to break chemical bonds. The effects of such type of radiation are thermal and non-thermal effects. Thermal effects are mediated by temperature changes induced by the fields. ELF fields arising from common devices are also unable to give thermal effects: frequency and intensity exposure is too weak. Non thermal effects are mechanisms that do not involve any macroscopic heating. These are related to

direct effects of electric or magnetic fields, particularly magnetic, on bodies(Kaune et al., 2002)².

The biological effects caused by EMFs depend on their intensity, frequency, volatility, physical characteristics and the effected tissue (Gholam et al., 2013)³. EMFs reach into the tissues, causing cellular dysfunctions, they lead to disorders such as insomnia, headache, and stress. These fields negatively affect blood biochemistry, digestive and circulatory systems, and increase the risk for cancer (Saadeldin., 2011)⁴. EM waves are to vibrate surface ions of cell membrane that disrupt the electric sensitive channels of plasma membrane that ultimately leads to cell malfunction. The heat of effects EM wave increases temperature of tissues, cells and increases blood flow, which leads to bleeding from capillaries is due to compression and destruction of tissue. Also, the thermal effects of EMFs, result disruption

proliferation of immature cells in the bone marrow, reduced red blood cells and white blood cells are reduced. EMFs, waves, electric current that stimulates nerves and causes tissue inflammation and decreased levels of interferon γ and the result is a weakening of the immune cell. EM waves in young tissues than in older tissues devastating effects due to radiation absorption rate, increasing the rate of cell division and low transfer rate due to lack of myelin nerve to leave a message (Gholam et al., 2013)³. The aim of our work is to investigate the effects of magnetic field of strength 50 Hz –3mTesla on some biochemical properties of rat tissue.

2. Material and Methods

Experimental animals and study design

The animals were exposed to a 50 Hz, 5mT (8hrs/day, 5days/week) homogenous magnetic field generated by four solenoids of 1500 turns each of electrically insulated 2.2 mm copper wire, wound around a copper cylindrical chamber of 17 cm external diameter as shown in Fig.1. Water was pumped in a copper jacket separating the wire winding and the chamber in order to keep the temperature of the chamber constant during the exposure period. The temperature of the flowing cooling water at the outlet of the jacket and the temperature inside the irradiation chamber were periodically measured through the use of thermocouple thermometer, which can give readings for the temperature variations within $\pm 0.1^\circ\text{C}$. There was no measurable difference in temperature between the room and the chamber. The actual current passing in the solenoids was about 1A. The animals were kept in special plastic cages that permit normal ventilation and daylight. The coils were connected to a variac fed from the mains (220 V and 50 Hz). The magnetic field exposure system was locally manufactured.

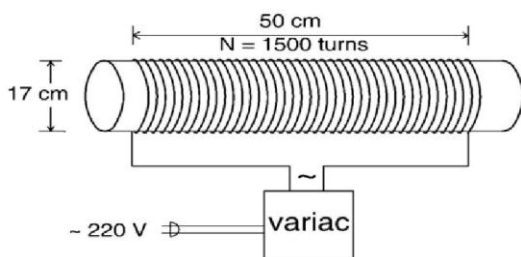


Fig. 1. Schematic diagram for exposure facility system hematological studies.

Collection of blood samples

The rats were slaughtered after the exposure periods, blood samples were taken at the end of exposure and collected on anti-clotting EDTA bottles (Harper & Rodwell, 1979)⁵, and these samples

were used in assessment of blood profile (Dilek et al., 2009)⁶.

Blood cell counts

The total red blood cell (RBC) counts were measured in millions per cubic millimeter ($\text{mil}/\mu\text{L}$) of blood. White blood cell (WBC) counts were measured in thousands per cubic milliliter ($\text{k}/\mu\text{L}$) of blood and blood platelet (PLT) counts are measured in thousands per cubic millimeter ($\text{k}/\mu\text{L}$) of blood. The blood is drawn in a test tube containing an anticoagulant (EDTA, sometimes citrate) to stop it from clotting as recommended by (Lewis et al., 2006)⁷.

Packed cell volume

Hematocrit values were determined by allowing the blood to flow into capillary heparinized tubes from decapitated rats then centrifuged in a hematocrit centrifuge at 1000g for 5 min. Mean corpuscular hemoglobin (MCH), mean corpuscular hemoglobin concentration MCHC and mean corpuscular volume (MCV), was calculated from RBCs count and hemoglobin count as follow (Cheesbrough, 2001)⁸.

$$\text{MCV} = (\text{hematocrit} / \text{RBCs count}) \times 10 \text{ fL.}$$

$$\text{MCH} = (\text{Hemoglobin concentration} / \text{RBCs count}) \times 10 \text{ picogram.}$$

$$\text{MCHC} = (\text{Hemoglobin content} / \text{Hematocrit}) \text{ g/dL.}$$

Hemoglobin content

Blood hemoglobin was determined using the Sigma reagent kits (Sigma Chemical Co., St. Louis, MO, USA) following their manual instruction. Hemoglobin was converted into cyanmethemoglobin by potassium ferricyanid and cyanid. Cyanmethemoglobin was measured colorimetrically where the intensity of color is proportional to the hemoglobin concentration. Twenty micro liters of the blood were added to 5 milliliters of diluted Drabkins solution. Standard was prepared by adding 20 micro liters of redistilled water to 5 milliliters of cyanmethemoglobin. The colors of the sample and standard were measured using Perkin-Elmer Lambda 1A UV-VIS spectrophotometer at 540 nm made in Taiwan (Mohammed, 2010)⁹.

Whole blood viscosity measurements:

A viscometer type Wells-Brookfield Cone/Plate DV-II manufactured by Brookfield Laboratories, Stoughton, MA) as shown in Fig (2). The temperature of the sample controlled by a refrigerated circulating fluid bath during measurement of the viscosity over a range of different temperatures. The cone/plate viscometer is a precise torque meter that is driven at discrete rotational speeds. The torque measuring system, which consists of beryllium-copper spring connecting the drive mechanism to a rotating cone, senses the resistance to rotation caused by the presence of sample fluid between the cone and stationary flat plate. A sample of fluid (blood in this

case) held between the rotating cone and a stationary flat plate. The resistance to the rotation of the cone produces a torque that is proportional to the shear stress in the fluid. This system is accurate to within $\pm 1.0\%$ of full scale range. Reproducibility is to within $\pm 2.0\%$. Working temperature range is from 0°C to 100°C . This method has been used to measure whole blood viscosity in units of centipoises (cp) over ranges of different temperature, at constant shear rate 100sec^{-1} in order to evaluate any changes in the membrane elasticity or in the ionic permeability and shape may cause change in the whole blood viscosity. The cup and cone were cleaned and dried thoroughly after the end of the experiment.



Fig.2: Brookfield viscometer and temperature control to control sample temperature during viscosity measurements.

Biochemical parameter

For the biochemical tests, about 2–3mL of blood sample depending on the rat weight was collected into a centrifuge tube without any anticoagulant. The centrifuge tube was left for about 15min to allow blood coagulation. Clear serum samples were then separated by centrifugation at 1000g for 20min. Serum samples were separated in glass tubes, labeled and stored in deep freezer at -50°C for different biochemical analysis for different biochemical assays. However, determination of enzyme activities was carried out on fresh serum samples. Serum aspartate aminotransferase (AST) is an enzyme belonging to the class of transferases. It is commonly referred to as a transaminase and is involved in the transfer of an amino group between aspartate and keto acids. AST activity is measured by using optimized ultraviolet-test according to International Federation of Clinical Chemistry and Laboratory Medicine (IFCC) (Thomas., 1998)¹⁰. Serum alanine aminotransferase (ALT) is a transferase with enzymatic activity similar to AST (Moss & Henderson.,1999)¹¹.

Whole blood was centrifuged twice at 3000 rpm for 10min in order to separate serum using a

biochemical autoanalyzer (Hitachi Automatic Analyzer 902, Roche), serum biochemical analysis was carried out. Kidney function was investigated through measuring urea and creatinine concentrations. Urea was determined according to the method reported by Fawcett and Scott (1960) using reagent kit obtained from Biodiagnostic, Egypt. Serum creatinine concentration was determined according to the method of Schirmeister et al. (1964) using reagent kits obtained from Biodiagnostic, Egypt.

Colorimetric Determination of Serum Total antioxidant:

The determination of the antioxidative capacity is performed by the reaction of antioxidants in the sample with a defined amount of exogenously provide hydrogen peroxide (H_2O_2). The antioxidants in the sample eliminate a certain amount of the provided hydrogen peroxide. The residual H_2O_2 is determined colorimetrically by an enzymatic reaction which involves the conversion of 3, 5, dichloro -2-hydroxyl benzenesulphonate to a colored product (Koracevice et al., 2001).

Statistical Analysis of Data:

The data were analyzed using one-way analysis of variance (ANOVA). Results were expressed as mean \pm standard error (SE) and values of $P > 0.05$ were considered non-significantly different, while those of $P < 0.05$ and $P < 0.01$ were considered significant and highly significant; respectively. F probability expresses the general effect between groups.

3. Results & Discussion

Measurements of blood parameters are the most important means to determine the health status of experimental animals. Blood and blood parameters are believed to be one of the primary particles that come in contact with EMF. Blood, comprised of ions are likely to react with induced EMF generated by EMF charges (Faiza et al, 2013)¹².

The present study reported that magnetic field exposure results in a significant decrease ($p \leq 0.001$) in the hematological constituents of blood such as hematocrit percentage and red blood cells count RBCs as compared to control group as shown in Fig (3&4). The depletion in the values of hematological parameters following magnetic field exposure may be attributed to (a) direct damage caused by lethal dose of radiation (b) due to over production of reactive oxygen species by EMFs interactions (Sisodia et al., 2013)¹³. Changes in hematological parameters may be attributed to inhibition by free radicals produced by EMFs interactions.

Hematocrit values were significantly decreased hence resulted in anemia. Usually, this type of anemia is mild and appears like the pseudo-anemia caused by sports. In this respect, regular physical activity,

especially extensive running and exercises increase iron loss causing mild iron deficiency. True iron deficiency can even occur especially when nutritional iron intake is insufficient and iron demand is increased.

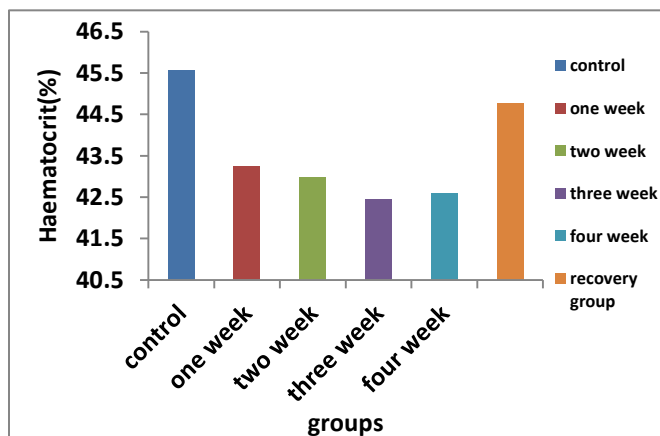


Fig.3. indicates hematocrit % for B₁, B₂, B₃, B₄ and B₅ as compared with control group A.

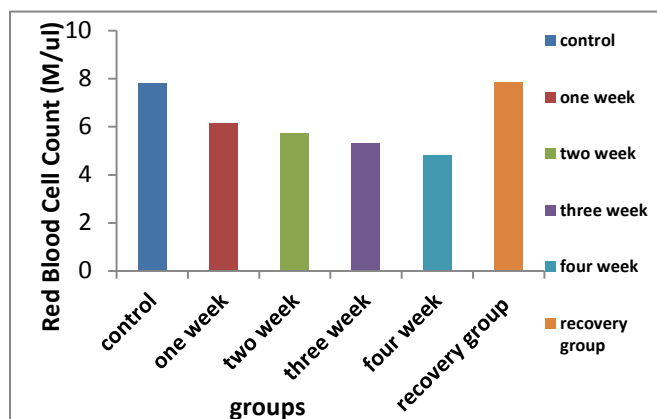


Fig. 4. indicates RBCs count for B₁, B₂, B₃, B₄ and B₅ as compared with control group A.

It was observed from the results of the reading recorded in Table.1. that the rate of hemoglobin decline were significant for all groups and not significant for recovery group B₅. The decline in haemoglobin and haematocrit after exposure to MF may be explained by the installation of hypoxialike

status which is probably resulting from the oxygen binding impairment of haemoglobin or iron metabolism disruption. Thus, exposure to MF decreased the serum iron level. This is in accordance with previous studies showing that exposure to electromagnetic field induced a decrease in blood.

The decreased of haemoglobin and red blood, after exposure to MF may be explained by the hypoxia-like status. However, the precise way in which MF induced hypoxia-like status has not yet been fully clarified. The hypothesis of an action of MF on the geometrical conformation of haemoglobin was reinforced by the fact that MF induced a prominent effect on the haemoglobin structure. High Significant increase in the WBCs for all groups following the end of exposure to MF indicated that exposure to magnetic field induced oxidative stress and free radicals generation in human blood platelets, producing a number of adverse effects and thus may lead to systemic disturbances in the human body (Mervat,2011)¹⁴. But there is an improvement for the recovery group B₅.

In the pine forest, Asteraceae was represented by nine species followed by Lamiaceae (7 spp.), Rubiaceae, (3 spp.), Fabaceae, Poaceae, Apiaceae, and Cyperaceae (2 spp. each) and remaining 14 families were represented by single species. Taxonomically, Asteraceae (with 9 genera) was the most diverse family followed by Lamiaceae, (with 7 genera), Apiaceae, Poaceae, Rubiaceae and Cyperaceae (with 2 genera each) and remaining 13 families were each represented by a single genus (Table 2).

The number of species varied spatially in both forests. In oak forest it varied from 15 (HT) to 30 (HB) and in pine forest from 12 (HT) to 23 (HB). Across the forests, maximum species were present in oak forest (at HB, 30) as compared to pine forest (at HB, 23). Species richness was higher (7.4) at HB and lower at HT (5.0) in oak forest. Similar pattern was found in pine forest, i.e., maximum species richness was at HB and minimum at HT (Table 3).

Table 1. RBCs, WBCs, platelets and Hemoglobin before and after exposure to magnetic fields

Parameters	RBCs Count (10 ⁶ /μl)	WBCs count (10 ³ /μl)	Platelets count. (10 ³ plate/ml ³)	Hemoglobin(g/dl)
A	7.81±0.67	7.3±0.14	712.71±29.9	14.3±0.11
B ₁	7.15 ±0.87 ^{VHS}	8.63±0.06 ^{VHS}	1121.67±63.4 ^{VHS}	14.4±0.13 ^{NS}
B ₂	5.72±0.76 ^{VHS}	10.429±0.91 ^{HS}	1052±12.88 ^{VHS}	13.85±0.17 ^{NS}
B ₃	4.81±0.87 ^{VHS}	12.09±1.19 ^{HS}	1149±13.3 ^{VHS}	14.35±0.38 ^{NS}
B ₄	4.12±0.36* ^{VHS}	12.614±0.93 ^{VHS}	757.86±13.29 ^{NS}	14.61±0.04 ^{NS}
B ₅	7.61±0.97 ^{NS}	7.1±0.06 ^{NS}	634±12.37 ^{NS}	14.686±0.32 ^{NS}

Values represented Mean ± standard error mean, NS, difference is not statistically significant between groups at level (p>0.05) S difference is statistically

significant between groups at level (p<0.01), HS difference is highly statistically significant between groups at level (p<0.001), VHS difference is very

highly statistically significant between groups at level ($p < 0.0001$).

After exposed rats to magnetic field, there is a significant effect in hemoglobin molecule structure due to damage/or broken of it as seen in Table.1. which also showed that there is an increased in the rate of haemoglobin after prolonged time periods when exposed to four weeks compared with the average rate in the control group, this means that these components of blood are broken due to irradiated by magnetic fields from technological devices which used daily. Data concerning hematological blood parameter levels for control and exposed groups are given in Tables.1. Results of the analysis are given as mean \pm standard deviation; also statistical differences were determined according to exposure period and groups. Indices of red blood cells give us a clear image of performance and efficiency of functional red blood cells, the concentration of hemoglobin, and describe the increase and decreased in the volume of red blood cells The most important of these indices that the average size of the red cells (mean cell volume), which helps in the diagnosis of some diseases, decreased in the size of the red cells showed in anemia. Fatayer reported that the mean corpuscular hemoglobin concentration (MCHC) index helps in the diagnosis of different types of anemia show Table.2.

The results showed a significant reduction in the measurements of the blood, hemoglobin and hematocrit (PCV), in addition to the number of indices of red blood cells are RBC, MCV, MCH, MCHC, this decline is an indication of different types of anemia, as well as leukemia (Mariam et al., 2012)¹⁵. While it was observed a significant decreased in the average number of white blood cells WBC as well as the proportion of lymphocytes LY and this decreased is accompanied with different cases of anemia, it is also evidence of bleeding which arises under the effects of radiation and increased the

temperature and resistance of the body. The increase or decreased in the percentage of lymphocytes associated with lymphatic leukemia, or inflammation of the lymph gland. Magnetic field exposure induced a decreased in Eosinophile, Hb, PCV and MCV levels.

The changes in lymphocytes may be due to the harmful action of electromagnetic field exposure that stimulates the haemopoietic system to release more lymphocytes causing changes in their number in the blood stream. Eosinophil level for rats of different groups showed non statistically significant changed as compared with control group A; however. There was a changed in lymphocytes, statistical difference in, monocytes, statistically significant increase in segmental neutrophil for all groups as compared with control groups. The exposure of rats to magnetic fields exhibited a general increase in WBCs (leukocytosis), and platelet count of albino rats. It has been shown that long-term exposure to magnetic fields can enhance the probability that mice carrying a lymphomagenic oncogene will develop lymphomas (Mohammed,2010)¹⁰. Still the question that can exposure to ELF-EMFs resulting from the production, distribution and use of electrical energy promote cancer or initiate any other health problems. This work is devoted to study the effects of such fields on some biophysical properties of red blood cells and tissues as a step forward to lay down safe limits and standards of exposure to these fields.

The effects of exposure to 50Hz-5mT magnetic field on red blood cells structural properties and tissues of albino rats were studied. Study of the effect of ELF-EMFs on living organisms is a complex problem, but it is of more interest to give insight penetrate the human body and act ions on all organs, altering the cell membrane potential and the distribution of ions and dipoles (Samira et al.,2008)¹⁶.

Parameters	Group(A)	Group(B ₁)	Group(B ₂)	Group(B ₃)	Group(B ₄)	Group(B ₅)
PCV (%)	45.571 \pm 0.665	43.243 \pm 0.87 ^S	42.986 \pm 0.755 ^S	42.457 \pm 0.87 ^S	42.6 \pm 0.362 ^{HS}	44.757 \pm 0.956 ^{NS}
MCV(FL)	82.43 \pm 0.12	51.83 \pm 0.62 ^{VHS}	71.53 \pm 0.43 ^{HS}	52.2 \pm 0.23 ^{VHS}	54.37 \pm 0.71 ^{VHS}	52.93 \pm 0.35 ^{VHS}
MCH (Pg)	27.56 \pm 0.31	17.56 \pm 0.45 ^{VHS}	25.8 \pm 0.36 ^S	17.8 \pm 0.66 ^{VHS}	18.59 \pm 0.65 ^{VHS}	16.99 \pm 0.25 ^{VHS}
MCHC(g/dl)	33.63 \pm 0.665	34.1 \pm 0.24 ^{NS}	33.71 \pm 0.60 ^{NS}	34.16 \pm 0.35 ^{NS}	33.31 \pm 0.65 ^{NS}	16.99 \pm 0.15 ^{NS}
LYM (%)	66.29 \pm 0.61	57.71 \pm 2.01 ^{HS}	74 \pm 1.56 ^{VHS}	50.29 \pm 2.66 ^{VHS}	69.71 \pm 0.47 ^{HS}	68.14 \pm 1.3 ^{NS}
MON (%)	2.57 \pm 0.20	7.67 \pm 1.17 ^{HS}	3.57 \pm 0.59 ^S	9.33 \pm 0.56 ^{VHS}	4.71 \pm 0.18 ^{VHS}	5.00 \pm 1.10 ^{VHS}
Seg (%)	25.43 \pm 0.65	27.67 \pm 1.33 ^{NS}	18.57 \pm 1.19 ^{VHS}	30.29 \pm 0.61 ^{VHS}	20.14 \pm 1.01 ^{VHS}	21.33 \pm 1.05 ^S
Eos (%)	1.14 \pm 0.14	3.86 \pm 0.34 ^{VHS}	1.57 \pm 0.20 ^{NS}	4.00 \pm 0.37 ^{VHS}	3.29 \pm 0.18 ^{HS}	2.00 \pm 0.31 ^S
Staff	2.29 \pm 0.20	1.17 \pm 0.18 ^S	1.29 \pm 0.18 ^{VHS}	2.00 \pm 0.31 ^{NS}	1.17 \pm 0.17 ^{HS}	1.33 \pm 0.21 ^{HS}
Neutrophils	29.14 \pm 0.40	32.14 \pm 1.01 ^S	24.43 \pm 1.4 ^{HS}	36.68 \pm 2.96 ^S	22.29 \pm 0.78 ^{NS}	28.43 \pm 1.21 ^{NS}

Values represented Mean \pm standard error, NS difference is not statistically significant between groups at level ($p > 0.05$) S difference is statistically significant between groups at level ($p < 0.01$), HS difference is highly statistically significant between groups at level ($p < 0.001$), VHS difference is very highly statistically significant between groups at level ($p < 0.0001$).

Exposures to ELF-EMF can alter the transcription and translation of genes (Gang et al., 2000)¹⁷, lead to generation of free radicals (ICRP- 60), influence cell proliferation rate and affect enzymes activities (Fadel et al., 2011)¹⁸. Table (2) Differential leukocyte count for blood before and after exposure to magnetic fields.

Mean corpuscular volume (MCV), mean corpuscular hemoglobin (MCH) decreased in response to electromagnetic field exposure compared to the control level. The changes in lymphocytes shown in table (2) may be due to the harmful action of magnetic field exposure that stimulates the haemopoietic system to release more lymphocytes causing an increase in their number in the blood stream. It has been shown that long-term intermittent exposure to magnetic field

can enhance the probability that mice carrying a lymphomagenic oncogene will develop lymphomas (Mohammed., 2010)¹⁰.

Changes in lymphocytes may be due to the harmful action of magnetic fields exposure that stimulates the hematopoietic system to release more lymphocytes causing an increase in their number in the blood stream. also have reported an increase in lymphocytes in cases of anemia, specifically macrocytic anemia, which arise under the influence of exposure to magnetic fields, increased temperature, and increased resistance to the body's immune system (Faiza et al.,2013)¹². Leukocyte analysis (total leukocyte count and the differential count (%)) for all groups is indicated in Fig 5.

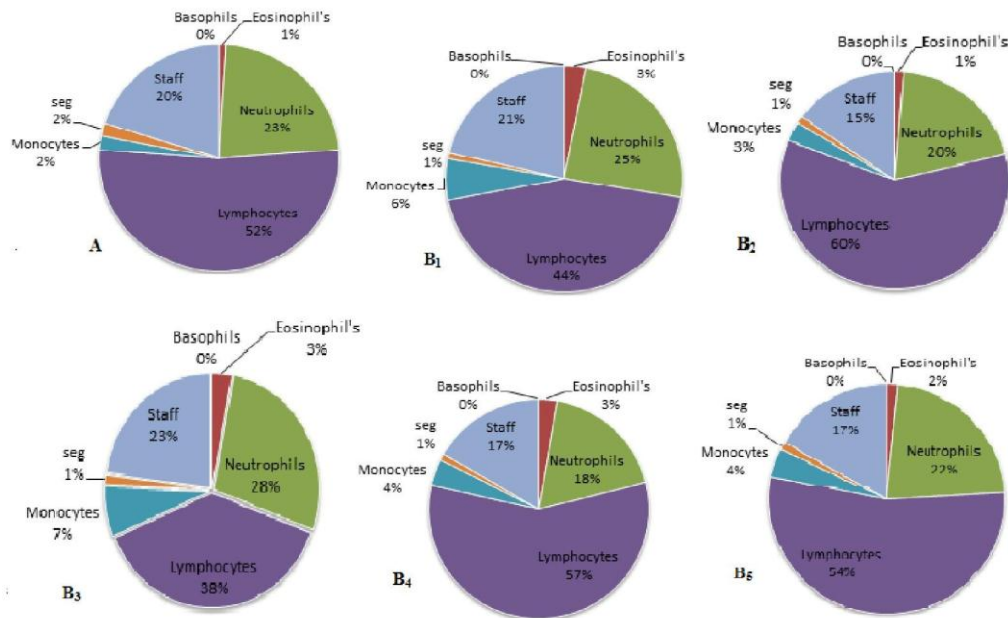


Fig.5. leukocyte analysis (total leukocyte count and the differential count (%)) for all groups.

From the results in Table (3) and Fig (6) it is clear that the viscosity of the blood was increased for animals of all groups as compared with control group A. The differences in viscosity demonstrate the effects of RBCs aggregation and deformability respectively.

The rat liver function was studied through analysis of alanine aminotransferase (ALT), aspartate aminotransferase (AST) and total protein (TP) after exposure to magnetic field. The same biochemical parameters have been evaluated in the blood serum of rats. The levels of ALT and AST were increased during the period of exposure to magnetic field. Also, a recovery was carried out after two weeks from stopping the exposure to magnetic field. The changes

in liver enzymes and total protein from blood serum analysis are shown in Table (4).

Table (3) illustrates the calculated values for the viscosity for samples of each group. *significant **very significant

groups	viscosity at room (Cp)± SEM
A	4.4±0.11
B ₁	5.45±0.12*
B ₂	7.56±0.21*
B ₃	7.85±0.65*
B ₄	8.71±0.21**
B ₅	8.32±1.34**

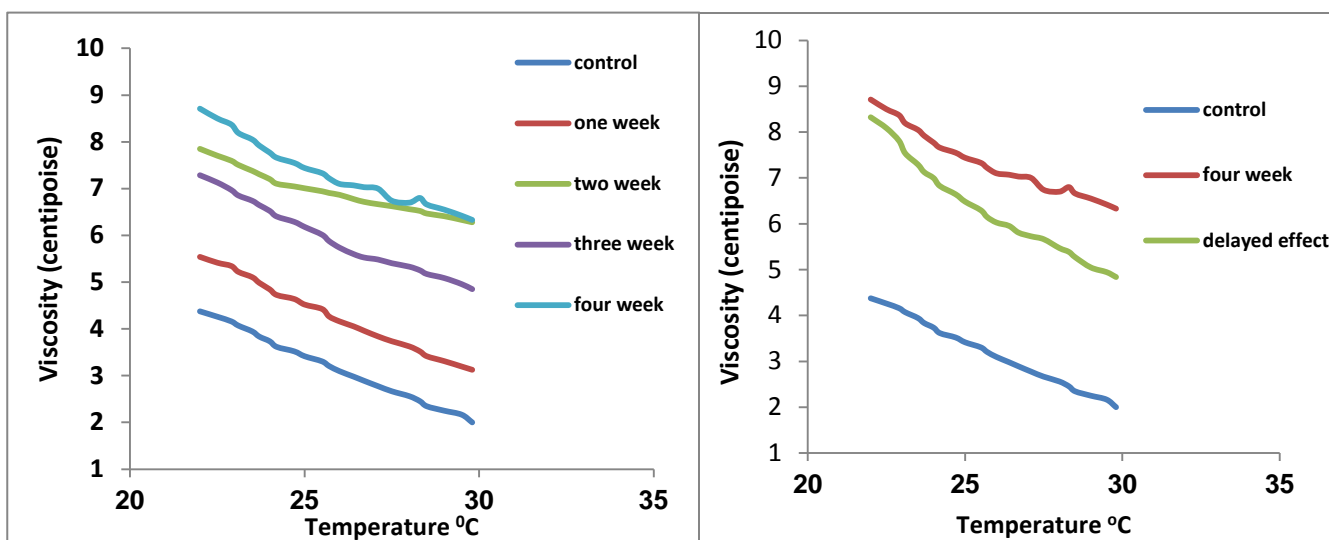


Fig. 6. Viscosity as a function of temperature for all groups B₁, B₂, B₃, B₄ and B₅ as compare with A

Table (4): Mean values of total protein TP, the alanine aminotransferase (ALT), aspartate aminotransferase (AST) of blood serum and kidney function for groups exposed to 50Hz-5mltesla and recovery (post exposed after 45 day). Values are the average of 7 experiments and P<0.001 as compared to values for the control group

groups	Total Protein Conc. (g/dl)	AST (u/l)	ALT (u/l)
A	6.233±0.08	66±4.43	46±1.46
B ₁	9.6±0.179 ^{VHS}	167.5±1.12 ^{VHS}	36.5±0.76 ^{VHS}
B ₂	6.7±0.037 ^{VHS}	109±4.38 ^{VHS}	173±38.44 ^{HS}
B ₃	8.0±0.358 ^{VHS}	150.67±2.2 ^{VHS}	46.67±2.79 ^{NS}
B ₄	7.63±0.168 ^{VHS}	195.17±4.85 ^{VHS}	174±5.8 ^{VHS}
B ₅	6.5 ±0.037 ^S	167.67±16.45 ^{VHS}	55.3±2.89 ^S

Values represented Mean ± standard error mean, NS difference is not statistically significant between groups at level (p>0.05), S difference is statistically significant between groups at level (p<0.01), HS difference is highly statistically significant between groups at level (p<0.001), VHS difference is very highly statistically significant between groups at level (p<0.0001)

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The changes in liver enzymes and total protein from blood serum analysis in Table (4) showed that MF produced alteration in biochemical parameters of the liver transaminases GOT and GPT which have been widely utilized in mammalian toxicology as biomarkers of specific organ dysfunction. In general the increase in transaminases activity is usually associated with hepatocyte damage. These results are in agreement with the results recorded by (Sihem et al., 2006)¹⁹. The authors studied the effects of sub-acute exposure to magnetic field on blood hematological and biochemical parameters in female rats and found that the serum GPT activity remained unchanged in treated rats, while GOT activity was increased, our present results agree

with observations obtained by many authors (Hudyma et al., 1990, Ibrahim et al., 2008, Shimanko et al., 1993)^{20,21,22}. The level of serum total proteins (TP) is significantly increased after exposure to MF.

These results were agreement with other findings reported by other works that in vivo exposure to a pulsed magnetic field at 1.5mT caused significant changes on plasma proteins in rats, difference in levels of plasma proteins were observed between the control groups of the two studies. This observation supports the hypothesis that the state of physiological equilibrium of a biological system is crucial to its response to a potentially effective EMF (Valberg et al., 1997)²³. The results showed that the exposure to time varying magnetic field induces EF and this in turn may cause large structural changes of the protein molecules imbedded in the cell membrane forming a new membrane conformation (Sahar et al., 2014)²⁴.

In this new conformation, the ions are able to pass through the membrane by binding temporarily with the protein molecule, thus “hopping” through the membrane (Watanabe et al., 1997)²⁵. showed that activities of GOT and GPT in the plasma, as indicator of hepatotoxicity, may alter the cell membrane potential and distribution of ions and dipoles. (Kula et al, 1999)²⁶ reported that the physicochemical action of an EMF consists of electron, ion, dipolar, macrostructural and electrolytic polarization.

The changes of liver enzyme and total protein from blood serum analysis ALT and AST are marker enzymes associated with liver parenchyma cells and levels are raised in acute liver damage. They are also present in red blood cells, heart cells, muscle tissue, pancreas and kidneys. When the body tissue or organ such as the heart or liver is diseased or damaged, ALT and AST are released into the blood stream (Samuel et al, 2013)²⁸. The significant increase in ALT activity indicates cytotoxic effect of non-ionizing radiation on hepatocytes inducing apoptosis and necrosis (Mohamed et al, 2009)³⁰.

The change of concentration of serum total protein elevation is generally due to either to loss of the fluid from the body (dehydration) or to increase in the globulin component. The change in the total protein concentration for exposed animal is in agreement also with the finding of the others. these finding could be attributed to the fact that cell membrane is a likely to be the primary site for cascade of events resulting in biological responses. These response could be due to different primary events such as: change in the diamagnetic properties of membrane phospholipid, variation of the induced membrane potential along the surface of the membrane, generation of primary free radicals that can from free radicals subsequently causing cell damage influences on the surfaces of the macromolecules with in cells, effects on the order in the membrane structure and charge movements which are critical enzymatic action, counter ion polarization and hence changes in the ionic environment of membrane embedded macromolecules and most evidently the effects on the membrane- mediated Ca^{++} process (Taha et al, 2003)²⁸. Considering the known or unknown harmful aspects of electromagnetic waves (EMWs), in this investigation as well, these waves lead to some changes in liver enzymes. In fact the main mechanism of EMWs is to vibrate surface ions of membrane cell that disrupt the electric sensitive channels of plasma membrane that ultimately leads to cell malfunction. The heat of effects electromagnetic wave increases temperature of tissues, cells and increases blood flow, which leads to bleeding from capillaries is due to compression and destruction of tissue. Also, the thermal effects of electromagnetic radiation, result

disruption proliferation of immature cells in the bone marrow, reduced red blood cells and white blood cells. EMWs, electric current that stimulates nerves and causes tissue inflammation. EMWs in young tissues than in older tissues devastating effects due to radiation absorption rate, increasing the rate of cell division and low transfer rate due to lack of myelin nerve to leave a message (Jelodar et al., 2013)³.

From Table (5) the results showed a significant increase in the concentration of urea and creatinine compared with the control group (non-exposed). The increase of the concentration of urea and creatinine causes chronic complications in some organs of the body (Muna., 2014)²⁹. EMF- exposure increased the serum creatinine and urea concentrations. These results may be due to the renal dysfunction associated with congestion of renal blood vessels, contracted glomerular tufts of some glomeruli and focal leukocyte aggregation by pathologic examination. Similarly, mice exposed to MFs (5T) for 48h increased significantly blood urea nitrogen, whereas creatinine levels remained unchanged. Contrarily, subchronic exposure of rats to MF (128mT, 1h/day for 30 days) had no effect on serum creatinine and urea levels. This discrepancy could be related to the difference of the intensity of the SMF and the exposure time (h/day) and duration (Mohamed et al, 2009)³⁰.

Table (5): Urea and creatinine concentration for kidney of all exposed groups as compared with control group

groups	Urea Conc. (g/dl)	Creatinine Conc. (g/dl)
A	23.5±1.853	0.632±0.023
B ₁	30±2.757 ^{NS}	0.627±0.002 ^{NS}
B ₂	27.58±1.781 ^{NS}	0.688±0.035 ^{NS}
B ₃	32.33±2.69 ^S	0.82±0.037 ^{HS}
B ₄	29.5±0.224 ^{HS}	0.666±0.0024 ^{HS}
B ₅	6.5 ±0.037 ^S	167.67±16.45 ^{VHS}

Values represented Mean ± standard error mean, NS difference is not statistically significant between groups at level (p>0.05), S difference is statistically significant between groups at level (p<0.01), HS difference is highly statistically significant between groups at level (p<0.001), VHS difference is very highly statistically significant between groups at level (p<0.0001).

Fig.7. Indicates to total antioxidant capacity in different experimental animals groups. Measurement of free radicals in the organisms is difficult because of their transient nature and the omplexity of available technique. Since the separate measurements of different oxidant and antioxidant molecules were not

practical, measurement of Total antioxidant capacity TAC has been suggested (Erel., 2004, Erel., 2005)^{31,32}.

It was proved in many experimental studies that electromagnetic fields with various physical parameters could intensify a generation of reactive oxygen species with subsequent disturbances of a balance between an intensity of oxidant processes and capacity of antioxidant defense system depending on the activity of antioxidant enzymes. This toxic phenomenon called oxidative stress, results stimulation of the process of membrane lipids peroxidation leading in a consequence to development of apoptosis and cell death (Impact of Selected Electromagnetic Fields on Prooxidant/Antioxidant Balance in Liver of Rats).

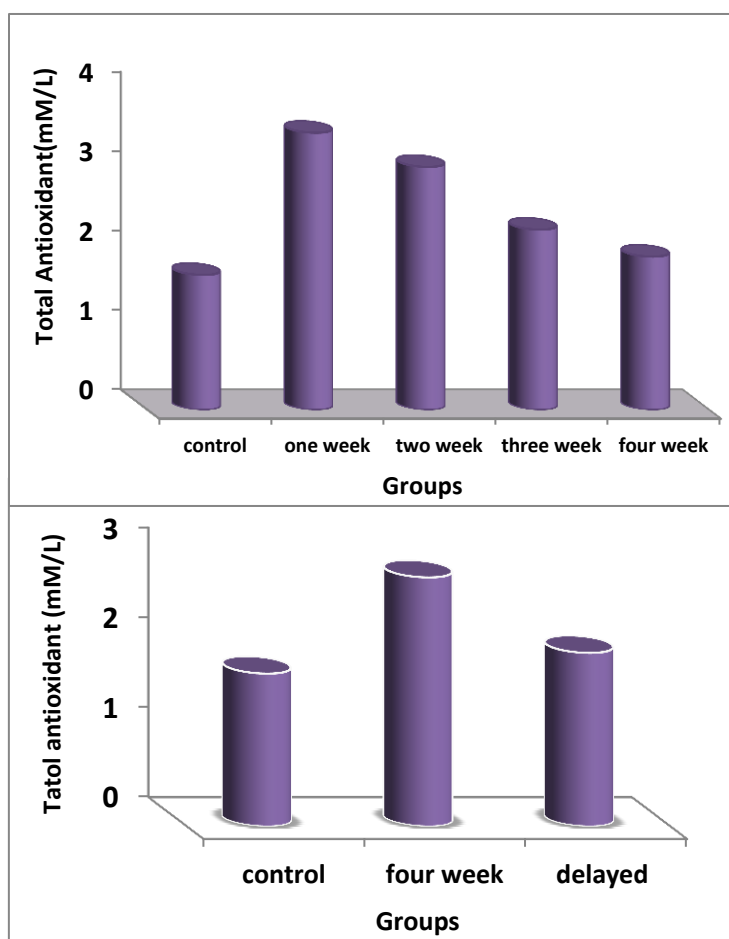


Fig. 7. Indicates to total antioxidant capacity in different experimental animals groups.

Conclusion

From the results of the CBC count, cell morphology and whole blood viscosity one may presume here that, the effect of prolonged exposures of the animals to 50Hz-5mT magnetic field resulted in changes in the mechanical properties, elasticity and ionic permeability of the RBCs membrane. These changes lead to the loss of the physiological mechanisms of the RBCs which will cause incomplete metabolic processes and as a result anemic diseases the result of post exposed group (recovery group) indicated that there is injury in the bone marrow due to exposure to the magnetic field which resulted in the generation of abnormal newly RBCs.

From the results of this work it may be concluded the following:

- Counting of blood in the medical examination for occupational workers is inadequate to evaluate the radiation hazards. It is necessary to measure the blood viscosity for radiation workers. Blood clot formation is not only indicated by measurements of the Thrombin level in blood, which is the running technique, but also by the loss of surface electric charge of the cells.

- It is necessary to review the dose limits recommended by the ICRP-60 for radiation workers based on the present findings.

- Prolonged exposures to strong magnetic fields are biological toxic.

- Exposure to 50 Hz-5mT magnetic field can cause anemic diseases which may cause heart problems.

- It is recommended not to allow buildings close or down to power-lines and special protocols for buildings permission should be done in a way that exposures to such fields are omitted.

- This study suggests that, in humans under investigation, the activities of liver enzymes GOT and GPT may increase and the conductivity may decrease by exposure to magnetic field generated during magnetic resonance imaging or nuclear magnetic resonance procedures.

- The decrease in conductivity due to field exposure and the recovery group not returned to the control value during the recovery period this is an indicator that there is no improvement in the liver state

Magnetic field exposure originated different metametabolic and hematological effects, which appeared to be related to the duration of exposure.

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