The Effect of Electromagnetic Field on Water and Fish *Clarias Garpienus*, Zagazig, Egypt

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Abstract: The aim of the present work is to study the effect of 50 Hz magnetic field of strength 2 mT on water and fish (*Clarias gariepinus*). In this work, the effect of extremely low frequency (ELF) and low intensity magnetic field on the biological systems was studied due to its existence everywhere in the environment from the electric appliances. Fish and water samples (from the tape water)were collected, half of the samples were used as control(pre-exposure) and the other half of the samples treated using electromagnetic field for two days. The physicochemical parameters of water samples were measured. Studies have been carried out on the blood and was investigated through measuring the dielectric relaxation in the frequency range 1 KHz to 4MHz at room temperature (for exposed and unexposed). From the data of the dielectric studies, we calculate each of the relaxation time, dielectric increment (ΔЄ),electric conductivity and Cole-Cole parameters of the (liver and kidney). The results of these studies indicated that the dielectric properties of (liver and kidney) exposed to 48hrs showed considerable changes in relaxation time, dielectric increment (ΔЄ), electric conductivity and Cole-Cole parameter of the (liver and kidney). The electromagnetic exposure caused an increase in temperature, a decrease in oxygen content and carbon dioxide, and no change in pH value, conductivity, salinity, chlorinity, alkalinity and total dissolved solids. In addition, it was observed that the trace element concentrations in water and the studied fish tissues decreased. The study shows that the electromagnetic field plays a major role in finding successful solutions for a lot of environmental problems as well as water pollution treatment.

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Key words: Electromagnetic field, water, fish, physicochemical parameters, water pollution treatment, dielectric increment.

1.Introduction

The biological effects of low frequency electromagnetic fields (EMF) have become a topic of considerable scientific interest scrutinized during the past two decades. Extremely low frequency magnetic fields (ELF-MF) are one of ubiquitous factors in the Earth's environment that may be emanated by various sources: geomagnetic fields, electric potential in the atmosphere or cosmic radiation. All the electrical devices that we use produce a low frequency electromagnetic field50Hz. All of the living organisms on the Earth, including plants, have been exposed to the natural electromagnetic field from the beginning of life and they have adapted to it. Numerous studies show an association of the internal electromagnetic field of plants with many physiological processes. The effects of weak magnetic and electromagnetic fields in biology have been intensively studied on animals **(Zwingelberg *et al*., 1993)**, micro-organisms **(Alvarez *et al*., 2006)** and humans **(Maes *et al*., 2000)**, but comparably less on plants **(Belyavskaya *et al*., 2004** and **Dhawi *et al*., 2009)** .From scientific literature, it is known that biological systems give different bio-responses to extremely low frequency magnetic field exposures at different frequencies and intensities **(Goodman *et al*., 1995)**. Various living organisms are differently affected from an extremely low frequency magnetic field, and these effects vary according to exposure conditions, genotype of organisms and the biological system **(Miyakoshi *et al*., 199 and Belyavskaya *et al*., 2001)**. In this way, several such studies have suggested that exposure to magnetic fields induces quite a variety of biological effects and, moreover, knowledge of the effects on living organisms is still not very clear.

Water pollution is regarded as one of the most critical environmental problems, as it causes change in water characters. The biological technique using the magnetic field to purify water was introduced. This technique is considered as a simple simulation of what happens in nature, as when water is subjected to a magnetic field and as a result, becomes more biologically active. The phenomenon of water treatment with an applied magnetic field has been known for many years and has been reported as being effective in numerous instances **(Balcavage *et al*., 1996)**. Magnetism sciences has developed and become more complicated, when its properties were found to have linkage with all solid, liquid and gaseous matters in addition to living organisms. **Johan-Sohaili *et al.* (2004)** explained that, magnetic technology is a promising treatment process that can enhance the separation of suspended particles from the sewage. **Tai *et al.* (2008)** observed that on subjecting water to magnetic field, it leads to modification of its properties, as it becomes more energetic and more able to flow which can be considered as a birth of new science called Magneto biology. He also pointed out that, it increases the percentage of nutrient elements like phosphorus and potassium. Magnetic wastewater treatment has been introduced to the chemical industry to remove heavy metals **(Tsouris, 2001)**. **Amiri and Dadkhah (2006)** found that changes in surface tension due to magnetic treatment, can be a key point in tracing impurities in water. Meaningful changes in surface tension of a liquid sample after a day can be a good indicator for the presence of physical and chemical changes in the sample. It was observed that magnetized water helps in dissolving minerals and acids by a higher rate than unmagnetized water, in addition to dissolving oxygen and increasing the speed of chemical reactions **(Moon and Chung, 2000)**.**Florenstano *et al.* (1996)** concluded that only the mineral content i.e., TDS (Total Dissolved Solids) that builds up after water is contacted with magnetic fields **(Florenstano *et al.*, 1996)**. Among different physical and chemical methods of water and wastewater treatments; magnetic methods attract a special attention due to their ecological purity, safety, simplicity and less operating costs. Alteration of physical and chemical properties of water-dispersed systems in the mode of magnetic treatment implies a certain influence of magnetic field on the structure of water and aqueous solutions. Previous researches made by several scientific societies has discovered that magnetic field can improve technological characteristics of the water, i.e. better salt solubility, kinetic changes in salt crystallization and accelerated colloidal coagulation. Magnetic field is known to create the asymmetry of hydrated shells due to its effect on water molecules situated around the charged particles (colloid). Exposure to magnetic field would lead to higher electro-kinetic movement among the colloid. This will definitely increase the probability of attracting particles to cloak with one another. The theory of magnetic field impact on technological processes for water treatment falls into two main categories, crystallization at magnetic water preparation and impurity coagulation in water systems **(Fadil *et al.*, 2001)**.

Experiments on animals revealed a favorable effect of magnetic fields in inflammatory processes, in radiation sickness and in experimental tumors. However, the therapeutic effect of electromagnetic field cannot be used successfully until we have a clear knowledge of the physiological and physicochemical mechanism of the biological influence of electromagnetic field. It was shown that electromagnetic field slows down the erythrocyte sedimentation rate, changes the number of leukocytes and phagocytal activity of leukocytes(PhAL) and their luminescence, Orientation of sickle-shaped erythrocytes of human blood across magnetic lines of force was discovered. It was shown that magnetic fields change osmotic processes in muscles, affect the permeability of the cellular membrane, and disturb the hydration ability of tissues in animals **(Kholodov, 1974)**.

Therefore, our aim in this study is to improve physical, chemical properties of water and fish tissues by subjecting samples to magnetic fields as a new, safe biological method for man and environment.

2.Material and Methods

1- The collected fish:

Clarias gariepinus (Fig. 1) is one of the most important freshwater fishes in Egypt. Total production of it in 2007 was about 31.9 thousand tons; i.e. it contributes about 17.5% of the total Nile catch in Egypt. It is grayish olive to olive brown to blackish above, white or grayish beneath **(Abdel-Hafez and El-Caryony, 2009)**.

Fig. (1): *Clarias gariepinus* **(Burchell, 1822)**.

2- Analytical procedures:

Water samples were taken from a tape water of Faculty of Science, Zagazig University, while fish samples were taken from Muweis canal, Zagazig city, Sharkia province, Egypt. The fish tissues (muscle, gills, kidney and liver) were taken from the fish samples, while the blood was taken as a serum after the centrifugation. Some of fish and water samples were subjected to the electromagnetic field (Fig. 2). The water samples were analyzed for water quality (chemical and physical characteristics of water). The trace element residues; zinc (Zn), lead (Pb), manganese (Mn) and aluminium (Al) were analyzed in water and fish blood (serum), muscle, gills, kidney and liver. Water samples for element analysis was treated with 1 ml of HCL in 500 ml sample to arrest microbial activities (for control samples).

Fig (2): The magnetic field exposure system was manufactured locally, in the faculty of Science Cairo University.

A-Water analysis:

I- Physico-chemical analysis of water:

***Temperature**:
Temperature was measured at the site of sampling, using a mercury thermometer of 0 to 50°C range.

*** Conductivity**:
Conductivity was measured according to **Gupta (2000)** by conductivity meter HI 98302 DIST 2.

***pH**:
pH was measured by using glass electrode pH-meter (Digital Mini-pH-Meter model 55).

*** Dissolved oxygen**:
Dissolved oxygen can be determined by Iodometric method (Winkler's method) as follow:

1- A glass stoppered bottle of known volume (125 ml) is filled with a water sample avoiding any bubbling.
No air should be trapped in bottle after the stopper is placed.
2- The bottle is opened and 1 ml of each manganous sulphate and alkaline potassium iodide solutions is added in it using separate pipettes.
3- A precipitate will appear. The stopper is placed and the bottle is shaked thoroughly. Sample at this stage
can be stored for a few days, if required.
4- 2 ml of sulphuric acid were added and shaked thoroughly to dissolve the precipitate.
5- Whole content or a known part of it, transferred gently (avoiding bubbling) to a conical flask. A few
drops of starch indicator were added, then titrated against sodium thiosulphate solution. The end point when initial blue color turns to colorless.

* If whole content is used for titration:

$$DO\ mg/l = \frac{V_1 N \times 8 \times 100}{V_2 - V_3}$$

* If a fraction of the contents is used for titration:

$$DO\ mg/l = \frac{V_1 N \times 8 \times 100}{V4\ (V_2 - V_3/V2)}$$

Where, DO = dissolved oxygen; V_1= volume of titrant (ml); N = normality of titrant (0.025); V_2 = volume of sampling bottle after placing the stopper (ml); V_3 = volume of manganous sulphate + potassium iodide solutions added (ml). The equivalent weight of oxygen is 8.
The value of DO in ml/l can be obtained by dividing the DO in mg/l by 1.43 **Gupta (2000)**.

*** Carbon dioxide (CO_2):**
CO_2 can be determined according to **Gupta (2000)**.

*** Total dissolved solids:**
For measurement of TDS the water sample is taken and filtered through glass fiber filter (Whatman, No. 42 or 44) to remove the suspended particles. 250 ml of clean filtrate is evaporated in an oven set at 180˚C±2˚C. This is obtained by the difference in the weights of the evaporation dish before taking the water sample and after complete evaporation of the same sample. The result is expressed in mg per liter i.e. if 250 ml sample is taken then the weight is multiplied by 4 to get per liter value. The formula to calculate the result is:

$$mg\ /L\ TDS = \frac{W2 - W1\ in\ mg \times 1000}{V\ (ml)}$$

where W1 and W2 are weights of empty dish and of dried TDS after oven drying in mg and V the volume of water sample in ml **(Ambasht, 1990).**

*** Chlorinity**:
For determining the chloride content of water the most common method is through the use of silver nitrate and is called Argento-metric methods. Chloride ion reacts with silver nitrate to produce white precipitate of silver chloride and at the end point the free silver ion (on totally precipitating chloride) reacts with chromate ion (of potassium chromate added to titration flask) to given reddish brown color of silver chromate. 0.2 N solution of silver nitrate prepared by dissolving 3.4 g of the chemical in distilled water and made up to 1 litre (stored in dark bottle in dark place) is taken in burette. To the 50 ml of the sample water 2 ml of potassium chromate 5% solution (5 g $K_2Cr_2O_7$ dissolved in 100 ml of water) is added and titrated upon the end point when red ting persists (**Ambasht**, 1990).

$$Cl^-\ mg\ /\ l = \frac{ml\ of\ AgNo_3 \times 1000 \times 35.5}{ml\ of\ sample}$$

Chlorinity (ppt) = gm. Chloride / litre.

*** Salinity:**
Salinity can be determined from the following formula **(Pipkin *et al.*, 1997)**:

Salinity g / l (ppt) = Chlorinity x 1.865.

***Alkalinity:**

Alkalinity can be determined according to **Gupta (2000)**.

II-Trace elements in water:

Twelve water samples were collected from a tape water for trace element analysis, half of which were exposed to the electromagnetic field, and the other put in cleaned bottles and stored until analysis was carried out. Trace element concentrations in water were determined by atomic absorption spectrophotometer (Perkin Elmer, 2280). The samples were prepared and analyzed in sequential for zinc, lead, manganese and aluminium according to **APHA (1985)**.

Trace element concentration ppm = reading of A. A. X volume of diluted solution/Volume of water sample

B-Fish analysis:

I- Trace elements in fish tissues:

Twenty one fish from *Clarias gariepinus* were collected from Muweis canal, Zagazig city for Trace element analysis. The collected fish were washed with deionized water, fourteen of which were exposed to elecromagnetic field and the others put in cleaned plastic bags and stored frozen until analysis was carried out. One gram of the prepared tissue sample was subjected to digestion by adding 5 ml of concentrated HNO_3in beaker and covered with a watch glass and gently heated till dryness on a hot plate, temperature not exceeding 300˚C. After cooling, 5 ml of concentrated sulphuric acid was added and the mixture was heated for one hour then allowed to cool down to the room temperature. Hydrogen peroxide solution (2 ml 30%) was added to the contents of the beaker and reheated. The latter treatment was repeated until a clear solution was obtained. The contents of the beaker were quantitatively transferred to 100 ml volumetric flask using deionized water and kept until measuring **(Abd El-Shafee, 2003)**.

Trace element concentration µg/g = reading of A. A. X volume of diluted solution/ Weight of sample (g.)

C- Blood parameters measurement:

Blood samples were collected in heparinized tubes after decapitation of the animals. Blood was collected in dry tubes for measurements of RBCs, WBCs count HCR, Hb, MCV, MCH, MCHC.

1- Red blood cell analytic parameters:

Red Blood cells are defined by three quantitative values:

I- The volume of packed red blood cells or HCR (Haematocrit).

II- The Hemoglobin (Hb).

III-The red blood cell concentration per unit volume (MCV).

*** Red Cell Count:**

It was important to count as many cells as possible, for the accuracy of the count was increased thereby; 500 cells should be considered the absolute minimum (**Dacie and lewis, 1984).**

Calculation:

$$\text{Red cell count (per1)} = \frac{\text{No. of cells counted}}{\text{Volume counted } (\mu l)} \times \text{dilution} \times 10^6.$$

I) Determination of Packed cell volume (HCR or Haematocrit value):

Above the red cells and not included in the figure for the HCR will be seen a grayish-red layer of leucocytes and above this, just below the plasma, a thin creamy layer of platelets. These comprise the "buffy coat" (**Dacie and lewis, 1984).**

II) Hemoglobin concentration (Hb):

We measured hemoglobin by using a photoelectric colorimeter. The cyanmethaemoglobin was used by hemoglobin-cyanide method (**Dacie and Lewis, 1984).** First, a collection of blood samples for determination of haemoglobin was made. Venous blood or free-flowing capillary blood added to any solid anticoagulant can be used. Measurements can be carried out on blood which has been stored at 4 °C for several days.

Calculation:

$$\text{Hb (g/l)} = \frac{\text{540 of test sample}}{\text{540 of standard}} \times \frac{\text{concentration standard (mg/l)} \times \text{dilution factor (e.g.201)}}{100}$$

III) Mean Corpuscular Volume (MCV):

The average volume of the red blood cells is a useful red cell index that is used in the classification of anemia and may provide insights into patho-physiology of red cell disorders. The MCV is usually measured directly with automated instruments but may also be calculated from the erythrocyte count and the HCR by means of following formula (**Greer *et al*., 2004).**

$$\text{MCV} = \frac{\text{HCR (1/1)} \times 1000}{\text{red cell count } (10^{12}/1)}$$

MCV was measured in femto liters (10^{-15} L). Using automated methods, this value was derived by dividing the summation of red cell volumes by the erythrocyte count.

IV) Mean Corpuscular Hemoglobin (MCH):

MCH was a measure of the average hemoglobin content per red cell. It may be calculated manually or by automated methods using the formula of (**Greer *et al*., 2004).**

$$MCH = \frac{\text{Hemoglobin (g/l)}}{\text{red cell count } (10^{12}/1)}$$

MCH was expressed in picograms (pg, or 10-12 g). thus, the MCH was a reflection of hemoglobin mass.

V) Mean Corpuscular Hemoglobin Concentration (MCHC):

The average concentration of hemoglobin in a giving red cell volume or MCHC may be calculated by the following formula (**Greer *et al*, 2004).**

$$MCHC = \frac{\text{Hemoglobin (g/dl)}}{\text{HCR (1/1)}}$$

The MCHC was expressed in grams of hemoglobin per deciliter of packed red blood cells. This represents measurement of Hb or the ratio of hemoglobin mass to the volume of red cells.

**** Leukocyte counts:**

Total counts by electronic methods **(Dacice and Lewis, 1984)**:

Differential leucocyte count (WBCs):

Differential leucocyte counts were usually performed on blood films which were prepared on slides by the spread or "wedge" technique (**Dacie and Lewis, 1984).**

Method:

The cells were counted using a 4 mm dry or 2.7 mm oil-immersion lens, in a strip running the whole length of the film.

***** Platelet counts:**

A platelet count was done for each sample using the formal-citrate method, and sometimes the ammonium oxalate method (**Dacie and Lewis, 1977 and Dacie and Lewis, 1991).**

Heparinized blood was diluted 1 to 100 in a freshly prepared formal-citrate solution (filtered before using), by adding 0.02 ml blood to 2 ml of the solution.

A neubauer counting chamber was filled with the suspension of the platelets using Pasteur pipette. The counting chamber was then placed in a moist chamber and left untouched for 20 minutes, and then platelets were counted.

Platelet count/cmm = N × 1000

N = Number of platelet counted in an area of 1 mm^2

D-Magnetic field exposure facility:

Equal volumes of the fishes were exposed to a homogenous magnetic field generated by a solenoid consisting of 320 turns from electrically insulated 2 mm cupper wire wound in a homogenous way around a cupper cylinder 2 mm thick, 40 cm diameter and 25 cm length. The cylinder wall was earthed to eliminate electric field components effects. The magnetic field generator by using the water cooling circulation around the cylinder was temperature controlled during the exposure period by using a water cooling shown in fig (2). The temperature during the exposure period was 37° C. The fish was put in the middle of the coil by using supports inside it to get a homogenous and higher magnetic field strength. The end of the coil was connected to variance fed from the mains (220Vpp, 50Hz).The field strength was adjusted by changing the voltage through the coil. The magnetic field exposure system was manufactured locally, in the faculty of science Cairo University. The magnetic field intensity was measured through the use of a magnetic flux meter type 4048 with probe T-4048.001. Manufactured by USA.

E- The dielectric measurements of organs tissue:

The liver and kidney organs were excised for the dielectric relaxation studies slides of each organs were placed in a special platinum cell has two squired electrodes with a length 1 cm each and area (A)(1x1cm^2) type (PW 950/60 manufactured by Philips) the distance between the electrodes was 1 cm (d) fig (3).

The relative permittivity $\varepsilon^{/}$ (dielectric constant), of the sample is defined as the ratio of the capacity measured with the sample to that measured by the cell in vacuum. The dielectric loss ($\varepsilon^{//}$) is the part of the energy of an electric field that is dissipated irrecoverably as heat in the dielectric.

The value of $\varepsilon^{/}$, $\varepsilon^{//}$ and the conductivity S sec^{-1} were calculated from measurements of the sample capacitance and resistance, The value of the relative permittivity (dielectric constant) ε' for the samples were calculated at each frequency from the measured values of its capacitance C (in Farad) through the equation

$$\varepsilon' = \frac{Cd}{\varepsilon_0 A} \qquad (1)$$

Where ε_0 is the permittivity of free space.

The dielectric loss (ε'') of each sample was calculated from its capacitance, C, and resistance, R (in Ohms), from the relation.

$$\varepsilon'' = \frac{\varepsilon}{2\pi RFC} \qquad (2)$$

The difference between the value $\varepsilon^{/}{}_{s}$ and $\varepsilon^{/}{}_{\infty}$ at low and high frequency is called “The dielectric increment $\Delta\varepsilon'$.

In case of biomolecules, the dielectric relation shows broader dispersion curves and lower maxima than those predicted by Debye model, and the $\varepsilon^{//}$

versus ἑ curves fall inside the semicircle. So (Cole-Cole, 1942) introduced a new parameter,α and modified Debye equation.
Moreover, the ac conductivity S (s^{-1}) was calculated from the equation

$$S = \frac{\sigma}{\varepsilon_0} = \omega\varepsilon'' = 2\pi f \varepsilon'' \qquad (3)$$

Whereε_0 is the permittivity of free space and σ is the actual conductivity.
Being the relaxation time, namely, the time at which the dielectric molecule has the ability to relax under the effect of the applied field and calculated from the relation

$$\tau = \frac{1}{2\pi f_c}$$

f_c being the critical frequency corresponding to the mid-point of the dispersion curve (or the frequency at the maximum loss) . The accuracy of the experimental set-up was about 1→3% in the whole frequency range investigated.

Fig (3): Hioki 3532-50 LCR meter.

F- Statistical analysis:

The statistical analysis was performed using the analysis of variance (ANOVA) to determine the differences between treatments mean at significant level of 0.05. Standard errors were estimated. All statistics were run on the computer using SPSS program. All graphics and tables were made by using Origin 8 and Microsoft word (2007). The methods used for analysis of the results were done according to **Bishop (1980).**

3.Results

A-Water analysis:

I- Physico-chemical analysis of water:

Comparing the average means of the same physicochemical parameters of water samples in the different exposure levels, the data recorded in table (1) showed a remarkable variations in it.

Table(1): The physico-chemical parameters (Mean ± SD) before and after the exposure to the electromagnetic field.

Magnetic field / Parameters	Pre-exposure	One day post-exposure	Two days post-exposure
Temperature (℃)	23.5±0.71^{ab}	27.5±0.71^{a}	29±0^{b}
Conductivity (ppm)	0	0	0
pH value	7.15±0	7.15±0	7.15±0
Salinity (‰)	30±0	30±0	30±0
Chlorinity(‰)	16.62±0	16.62±0	16.62±0
Total dissolved solids (ppt)	0.2±0	0.2±0	0.2±0
Dissolved oxygen (ppm)	0.25±0.07071^{a}	0.2±0	0.1±0^{a}
Carbon dioxide (ppm)	16.5±2.12^{ab}	2.5±0.71^{a}	2.5±0.71^{b}
Akalinity (ppm)	109±1.41	109±1.41	109±1.41

* Data are represented as mean ± SD, (n = 12).
**Means with the same letters in the same row are significantly different (*p*<0.05), using ANOVA.

II-Trace elements in water:

Comparing the average concentrations trace elements in the different exposure levels, the data recorded in table (2) and fig. (4) showed remarkable variations in trace element concentrations in water samples. The concentrations had the order: MN>Al >Pb> Zn.

Table(2): The trace element concentrations (Mean ± SD) of water before and after the exposure to the electromagnetic field.

Magnetic field / Parameters	Pre-exposure	One day post-exposure	Two days postexposure
Lead (Pb)	0.29±0.01	0.225±0.06	0.175±0.04
Aluminium (Al)	0.97±0.03^{a}	0.915±0.01^{b}	0.66±0.1^{ab}
Manganese (Mn)	2.735 ±0.78	2.09±0.45	1.875±0.53
Zinc (Zn)	0.15 ±0^{a}	0.085±0.01^{a}	0.06±0^{a}

* Data are represented as mean ± SD, (n = 12).
**Means with the same letters in the same row are significantly different (*p*<0.05), using ANOVA.

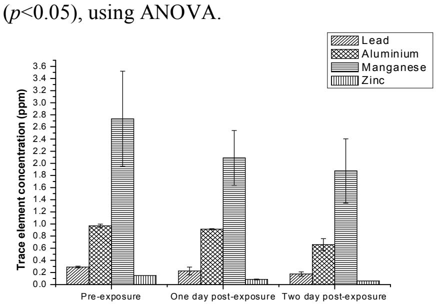

Fig (4): The trace element concentrations(Mean ± SD) in water before and after the exposure to the electromagnetic field.

B-Fish analysis:

I- Trace elements in fish tissues:

Comparing the average concentrations of trace elements in the different exposure levels, table :[3]and figs. : [5 -9] showed variations between trace element concentrations in fish tissues. The concentrations had the order: Mn>Al >Zn >Pb.

Table(3): The trace element concentrations(Mean ± SD) in some fish tissues before and after the exposure to the electromagnetic field.

Magnetic field / Parameters		Pre-exposure	One day post-exposure	Two days post-exposure
Blood	Lead (Pb)	3.47 ± 1.23	2.89 ± 0.21	2.73 ± 0.17
	Aluminium (Al)	8.96 ± 0.08	7.88 ± 0.95	7.62 ± 1.23
	Manganese (Mn)	94.35 ± 27.65[ab]	12.605 ± 3.27[a]	10.45 ± 6.05[b]
	Zinc (Zn)	3.77 ± 0.01[a]	3.005 ± 0.19 [a]	2.53 ± 0.11[a]
Muscle	Lead (Pb)	0.15 ± 0.01[a]	0.14 ± 0.00	0.13± 7.07E-4[a]
	Aluminium (Al)	1.14 ± 0.06[ab]	0.93± 0.06[a]	0.81± 0.01[b]
	Manganese (Mn)	13.02± 0.90[ab]	10.73 ± 0.08[a]	9.08 ± 0.13[b]
	Zinc (Zn)	1.54± 0.04[a]	1.43± 0[a]	0.78 ± 0.01[a]
Gills	Lead (Pb)	0.18± 0.00a	0.17± 0.00[b]	0.13± 0.02[ab]
	Aluminium (Al)	1.54 ± 0.11	1.39± 0.01	1.33± 0.06
	Manganese (Mn)	32.25 ± 3.46	31.91 ± 3.81	23.54± 1.90
	Zinc (Zn)	1.88 ± 0.04 [ab]	0.92± 0[a]	0.86± 0.04[b]
Liver	Lead (Pb)	0.21 ± 0.01[a]	0.18 ± 0.00	0.14 ± 0.02[a]
	Aluminium (Al)	2.21± 0.81	1.47 ± 0.06	1.34± 0
	Manganese (Mn)	36.86± 0.28	34.75 ± 1.06	30.96± 5.20
	Zinc (Zn)	2.20 ± 0.25	1.99± 0	1.89± 0.04
Kidney	Lead (Pb)	0.25± 0.01	0.24± 0.00	0.20± 0.04
	Aluminium (Al)	2.8± 0.01 [a]	2.42 ± 0.06	1.81± 0.49 [a]
	Manganese (Mn)	69.35± 5.73[a]	50.5± 2.12	43.4± 8.50[a]
	Zinc (Zn)	2.375 ± 0.01[a]	2.04 ± 0[a]	1.99 ± 0[a]

* Data are represented as mean ± SD, (n = 21).

**Means with the same letters in the same row are significantly different (*p*<0.05), using ANOVA.

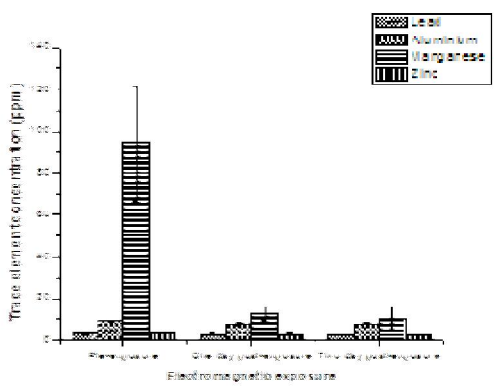

Fig (5): The trace element concentrations(Mean ± SD) in blood before and after the exposure to the electromagnetic field.

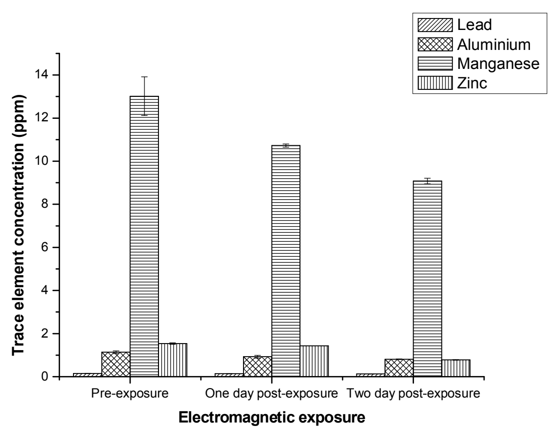

Fig (6): The trace element concentrations(Mean ± SD) in muscle tissue before and after the exposure to the electromagnetic field.

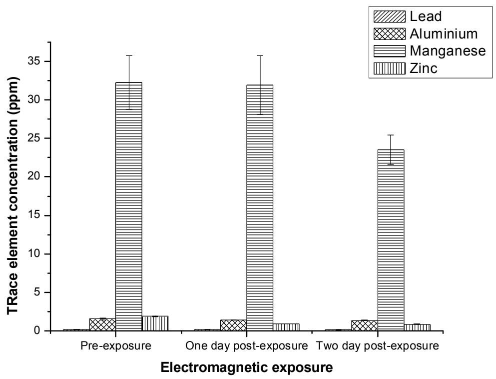

Fig (7): The trace element concentrations(Mean ± SD) in gills before and after the exposure to the electromagnetic field.

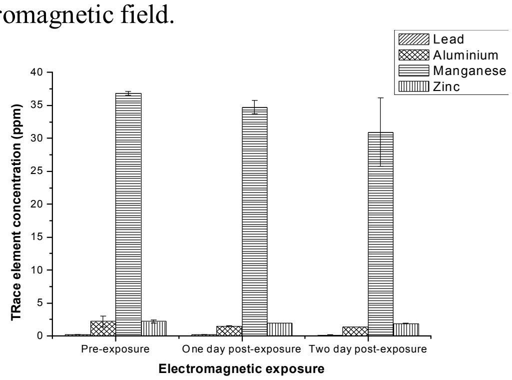

Fig (8): The trace element concentrations(Mean ± SD) in kidney before and after the exposure to the electromagnetic field.

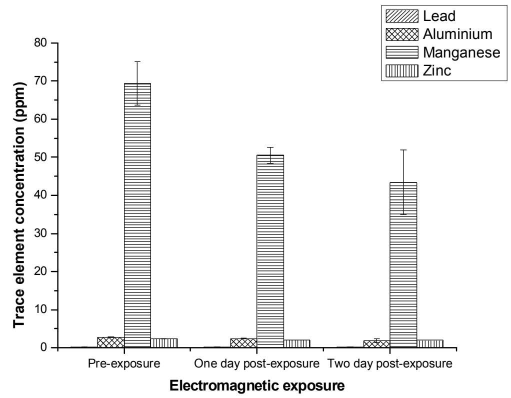

Fig (9): The trace element concentrations(Mean ± SD) in liver tissue before and after the exposure to the electromagnetic field.

C- The blood parameters:

The changes in the blood parameters such as, RBC, WBC. Count, HB, MCV, MCH, MCHC and HCR were measured for the control and exposed, the results indicated slight changes in all the blood parameters as shown in the table (4).

Table (4): the effect of 50 Hz magnetic field of strength 2 mT on Red Blood Cell (RBC) count, blood haemoglobin (Hb), Mean Corpuscular Haemoglobin (MCH), White Blood Cell (WBC) count, Packed Cell Volume (PCV) and plasma haemoglobin (Hb) on fish.

FISH No.	WBCs.per mm^3	RBCs	HB	HCR	MCH	MCV	MCHC
Control	12448±702	5.7±0.14	12.04±0.27	31.2±0.57	26.5±0.84	54.83±1.28	38.7±0.99
Exposed (24hrs.)	13698±534.6	5.24±0.25	10.8±0.38	28.1±0.9	26±0.43	54.16±1.87	38.5±0.63
Exposed (48hrs.)	17373±595.4	5.58±0.07	12.8±0.13	33.8±0.59	26.4±0.64	60.52±1.2	38.08±0.83

D- Dielectric relaxation Measurements:

The values of relative permittivity ε′, dielectric loss ε″ and conductivity S had been calculated through using the equations (1), (2) and (3) and then plotted. Figures (10a), (11a),(12a),(13a), (14a) and (15a) illustrate the variation of the relative permittivity ε′, dielectric loss ε″ plotted on the left Y axis and conductivity S on the right Y axis as a function of log frequency for volumes A_1, B_1, A_2, B_2, A_3, B_3 respectively. Figures (10b), (11b), (12b), (13b), (14b) and (15b) illustrate the Cole-Cole plot of the variation of the relative permittivity ε′ versus dielectric loss ε″ for the same volumes.

It is clear from the figures that the permittivity passes through a dielectric dispersion and also the conductivity S passes through the conductivity dispersion that resulted from the interfacial polarization.

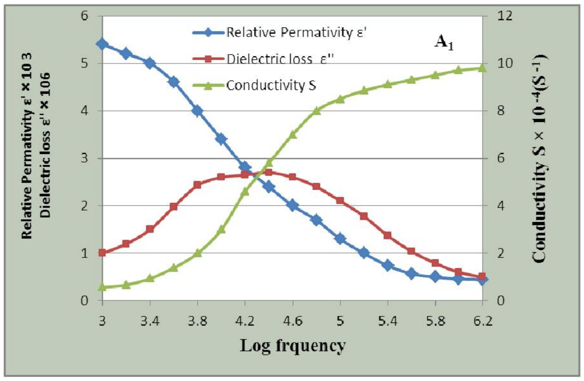

Fig.(10a): The dielectric Properties for control (liver) from fish

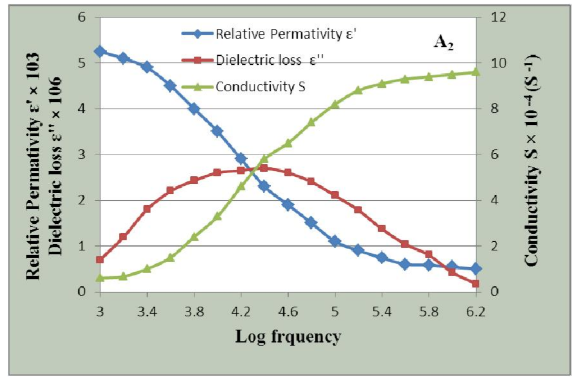

Fig.(12a): The dielectric Properties for liver from exposed (24h.)

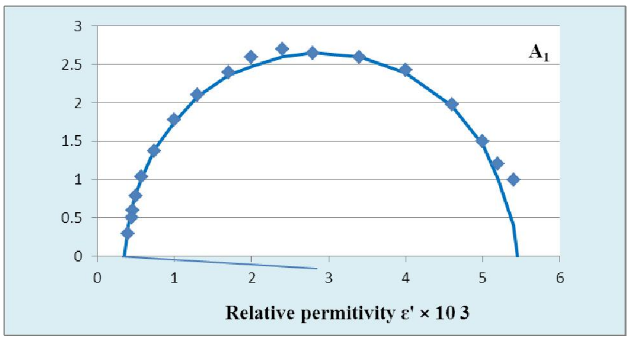

Fig.(10b): The Cole- Cole plot for control (liver)

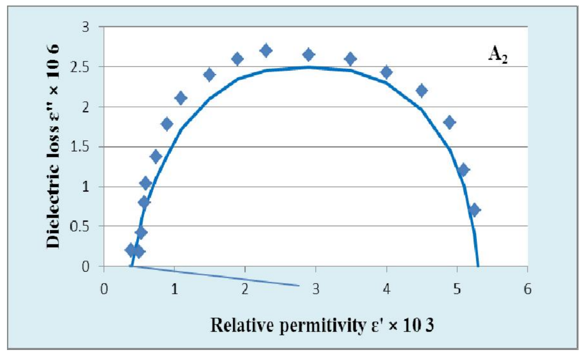

Fig.(12b): The Cole- Cole plot for liver from exposed (24h.)

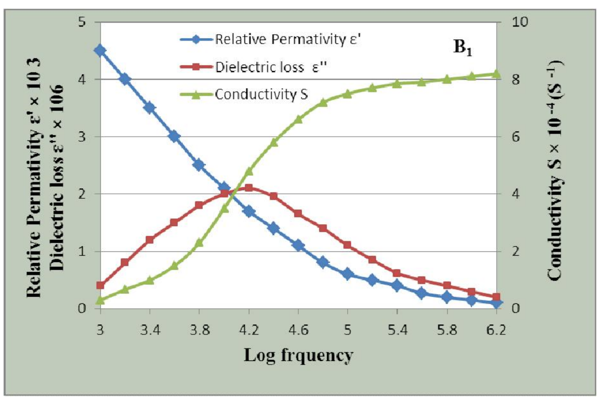

Fig.(11a): The dielectric Properties for control (kidney) from fish

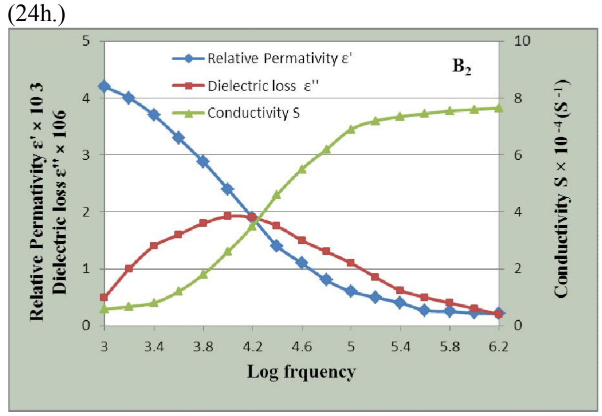

Fig.(13a): The dielectric Properties for kidney from exposed (24h.)

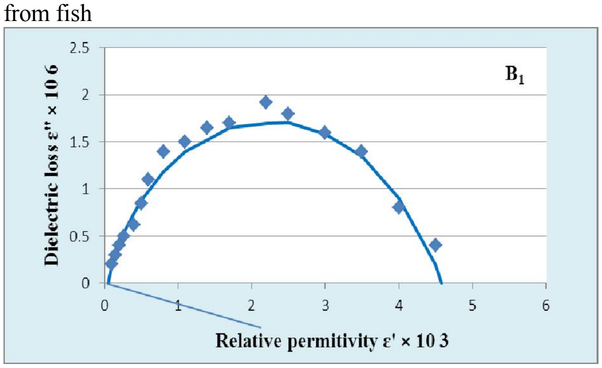

Fig. (11b): The Cole- Cole plot for control (kidney)

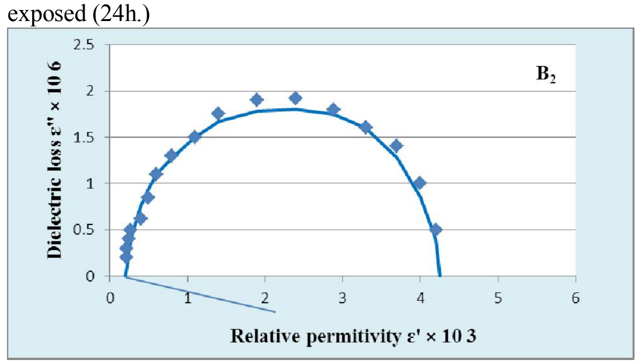

Fig.(13b): The Cole- Cole plot for kidney from exposed (24h.)

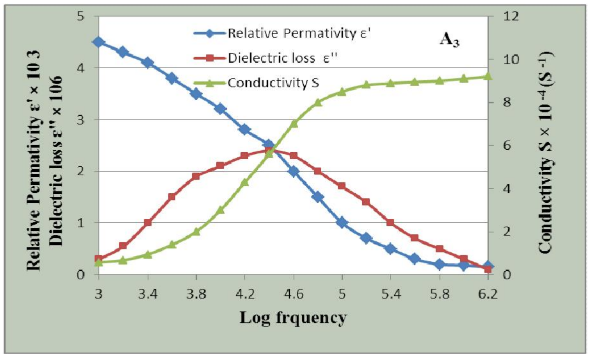

Fig.(14a): The dielectric Properties for liver from exposed (48h.)

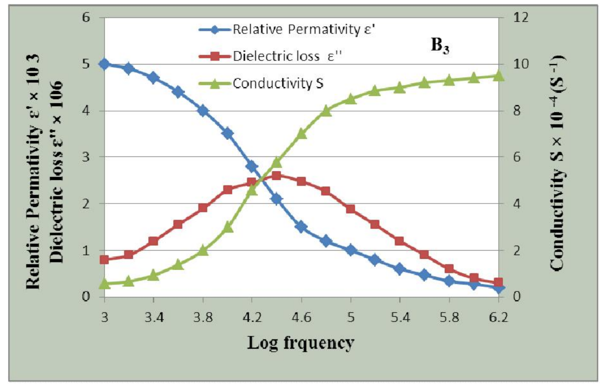

Fig.(15a): The dielectric Properties for kidney from exposed (48h.)

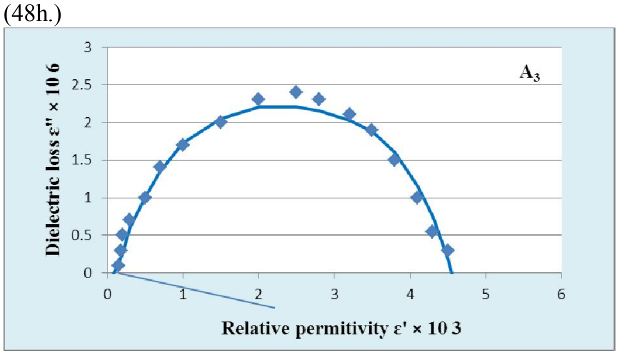

Fig.(14b): The Cole- Cole plot for liver from exposed (48h.)

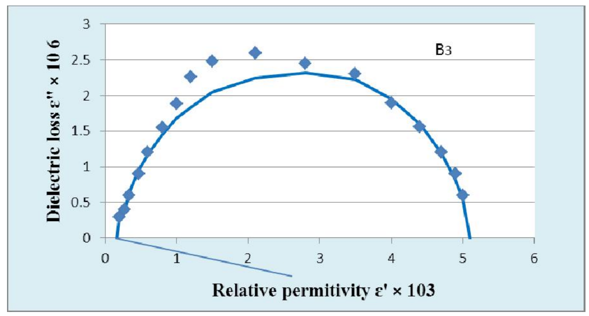

Fig.(15b): The Cole- Cole plot for kidney from exposed (48h.)

The results showed pronounced reduction in the values of the dielectric increment ($\Delta\varepsilon'$) and the conductivity (S) and an increase of the values of the relaxation time (τ), the average molecular radii (r) and the Cole-Cole parameter (α) for exposed volumes relative to unexposed.

Table (5): The dielectric Properties for control (liver, kidney) from fishes.

Samples	Volumes	$\Delta\varepsilon' \times 10^{6}$	$S \times 10^{-8}$ (s^{-1})	$\tau \times 10^{-4}$ (sec)	$r \times 10^{-8}$ (m)	α
Liver	A_1	5.1 ± 0.01	9.8 ± 0.022	2.25 ± 0.01	4.13 ± 0.04	0.05
kidney	B_1	4.79 ± 0.05	8.2 ± 0.06	4.49 ± 0.05	5.23 ± 0.075	0.15

Table (6): The dielectric Properties for exposed (24hrs.) for (liver, kidney) from fishes**.**

Samples	Volumes	$\Delta\varepsilon' \times 10^{6}$	$S \times 10^{-8}$ (s^{-1})	$\tau \times 10^{-4}$ (sec)	$r \times 10^{-8}$ (m)	α
Liver	A_2	4.1 ± 0.02	8.9 ± 0.021	2.01 ± 0.02	3.13 ± 0.03	0.04
kidney	B_2	3.29 ± 0.01	6.2 ± 0.07	3.49 ± 0.01	4.23 ± 0.055	0.11

Table (7): The dielectric Properties for exposed (48hrs.) for (liver, kidney) from fishes.

Samples	Volumes	$\Delta\varepsilon' \times 10^{6}$	$S \times 10^{-8}$ (s^{-1})	$\tau \times 10^{-4}$ (sec)	$r \times 10^{-8}$ (m)	α
Liver	A_3	4.62 ± 0.04	8.23 ± 0.02	2.12 ± 0.025	3.30 ± 0.07	0.091
kidney	B_3	3.04 ± 0.020	6.65 ± 0.115	1.52 ± 0.02	4.39 ± 0.14	0.15

4.Discussion

Low frequency magnetic fields are widely applied in electrical appliances and different equipment such as television sets, computers and kitchen appliances. Recently, low frequency magnetic field has been considered to be a therapeutic agent and it has started to be more and more commonly used in medicine. Electromagnetic fields have adverse effects as a result of widespread use of electromagnetic energy on biological systems.

In recent years a large number of multidisciplinary investigations led to the increasing awareness of the existence of multiple effects in biological systems. The responds of acute cardiovascular system to an electric and magnetic field is still being analyzed. Several experimental and biological studies have dealt especially with increased incidence of various types of cancer. *In vivo* and *in vitro* investigations claim that extremely low-frequency magnetic field produced a genotoxic effect.

The results presented in this work concerned with the induced changes in the structural and characteristic behavior of fish resulting from its exposure to magnetic fields are of biophysical and medical importance and interest. This data when piled together can lead to important conclusions which may be of great importance for evaluating the benefits as well as the hazards from the exposures to low frequency low level magnetic fields. The data of the dielectric relaxation as given in tables: [5-7] may spotlight the changes for (liver, kidney,) from fishes and changes in blood parameter.

Form tables (6 -7) it's clear that there are decreasing in the values of dielectric increment $\Delta\varepsilon$, conductivity S, relaxation time (τ) for the exposed fish relative to the unexposed fish. Moreover, the magnetic field at such frequency (50Hz) may also be in resonance with one of the metabolic activities that might occur through fish membrane and cause the changes in all their biological properties. To get better understanding of the interaction mechanism of magnetic field with biological systems and understanding of the bioelectrical signals resulting from biological system during metabolic activity is required. **Mohamed *et al.* (1997)** reported that the bioelectrical signals from the microorganism normally were carried out through bending of their cellular membranes which generate electric impulse through phenomena known as flex electricity. The amplitude and the frequency of these impulses depend on the amount and frequency of bending.

Water pollution is due to the change in physical, chemical and biological properties which is either directly or indirectly caused by human activity and their derivatives. It is thought that modifications to the properties of solutions through the magnetic field changes in the molecular structure of liquids, polarization, resulted from arrangement of particles and finally from changes of the electric potential **(Lebkowska *et al*., 1991; Szczypiorkowski, 1995 and Krzemieniewski *et al*., 2002)**.

Temperature:

Water temperature is one of the most influential environmental factors affecting both the metabolism and growth of fish and their body composition **(Weather and Gill, 1983; Herzing and Winkler, 1986)**.

In this study, the fluctuation in water temperature in the various examined levels was caused by electromagnetic exposure which leads to an increase in it **(Shatalov, 2009)** and this range of water temperature was favorable for fish according to results obtained by **Ayoola and Kuton (2009)**; **Owei and Ologhadien (2009)**; **Rahim *et al.* (2009)**; **Sithik *et al.* (2009)**.

Conductivity:

In the present study, the conductivity at all the examined levels was not changed and this disagreed with **Alkhazan and Sadddiq (2010**) who stated that it decreased after the electromagnetic exposure for 30 days and this may be related to the period of exposure (2 days**).**This may be attributed to the short period of electromagnetic exposure and weak intensity of the electromagnetic field, and it is beneficial as the electromagnetic exposure does not affect the physical properties of water in the short exposure.

pH- value:

In the present study, the pH values at all the examined levels were always at the neutral side and there was no effect of the electromagnetic field on it. The increase in summer can be related to increase in primary production which leads to increase of photosynthesis that involves the uptake of free carbon dioxide from the water and precipitation of calcium carbonate **[Saeed (2000); Islam (2007), Pandey and Tiwari (2009)** and **Khater (2010)]**. It is disagreed with that of **Alkhazan and Sadddiq (2010**) who stated that the pH- value increased with electromagnetic field and **Shatalov (2009)** who observed a decrease in pH value with electromagnetic exposure. It may be attributed to the short period (2 days) of the exposure in this study. The data obtained in this study indicate that the pH at all the study levels lies within the favorable limits (6.2-8.3) needed for the growth and survival of fish and comply with results of **Adeyemo *et al*. (2008); Korai (2008) and Pandey and Tiwari (2009).**

Dissolved oxygen (DO):

The decrease in co_2 after the electromagnetic exposure in the study is agreed with that of **Shatalov (2009).** From the obtained data, it is clear that DO content at all study levels lied below the limit (>5 mg/l) that satisfy the needs of successful fish production as reported by **ANZECC (2000); Saeed (2000); Ayoola and Kuton (2009); Pandey *et al.* (2009) and Sithik *et al.* (2009).** This is due to the type o water (tap water).

Carbon dioxide (CO_2):

The decrease in CO_2 after the electromagnetic exposure in the study is agreed with that of **Shatalov (2009)** and **Alkhazan and Sadddiq (2010).** From the obtained data, it is clear that CO_2 content at all study levels lied below the limit (>5 mg/l) that satisfy the needs of successful fish production as reported by **ANZECC (2000); Saeed (2000); Ayoola and Kuton (2009); Pandey *et al.* (2009) and Sithik *et al.* (2009).**

Salinity& Alkalinity:

From the present data, it is clear that the salinity and alkalinity contents were not changed in the study levels and this disagreed with **Ni´ am *et al.* (2006)** and **Alkhazan and Sadddiq (2010**), and may be related to the short period of the electromagnetic exposure and weak intensity of the electromagnetic field. Although, the range of salinity in the study(<3 mg/l)was suitable for fish growth and survival, the range of alkalinity(20 mg/l) was not recommended, as mentioned by **ANZECC (2000); Ayoola and Kuton (2009)**; **Sithik *et al.* (2009)**.

Total dissolved solids (TDS):

From the data reported in this study, it is clear that the values of TDS were not changed after the exposure to the electromagnetic field and this disagreed with **Ni´am *et al.* (2006)** and may be related to the short period of the electromagnetic exposure.

Heavy metals in water:

Zinc (Zn):

The mean concentrations of Zn in this study were below the legal limits (3 mg/l) recommended by **WHO (2008)** at all the study sites. These results are nearly similar to those obtained by **İncekara (2009)**; **Miclean *et al.* (2009) and Khater (2010)**, whereas they are lesser than those reported by **Akoto and Adiyiah (2007); Obasohan (2007)**; **Frankowski *et al.* (2009)**. Moreover, the mean concentrations of Zn decreased after the exposure to the electromagnetic field.

Lead (Pb):

The mean levels of Pb obtained here were higher than legal limits (0.01mg/l) reported by **WHO (2008)** in water samples collected from all the study sites. The concentrations of Pb in the present study were lesser than those recorded by **Abd El-Kader (1994); El-Safy (1996); Khalil and Hussein (1997); Ilyas and Sarwar (2003)**; **Gabriel *et al.* (2006)**; **Muwanga and Barifaijo (2006)**; **Abulude *et al.* (2007)** and nearly similar to those obtained by **Haggag *et al.* (1999); Saeed (2000); Yousafzai (2004)**; **Awofolu (2006); Gabriel *et al.* (2006); Frankowski *et al.* (2009)**. However, the electromagnetic exposure for 2 days decreased the mean concentrations of Pb.

Manganese (Mn):

The mean levels of Mn obtained here were higher than the legal limits (0.4 mg/l) recommended by **WHO (2008)** in the water samples. Comparative Mn levels were recorded by **Akoto and Adiyiah (2007); Obasohan(2007).** However, high concentrations of Mn were recorded by **İncekara (2009)**; **Abdul Ghaffar *et al.* (2009) and Khater (2010)**. Also, as in Pb, the electromagnetic exposure for 2 days decreased the mean concentrations of Mn.

Aluminium (Al):

In this study the mean Al concentrations were higher than the permissible levels (0.2 mg/l) recommended by **WHO (2008)** at all the study sites. The concentrations of Al in the present study were lesser than those recorded by **Awofolu (2006)**; and nearly similar to those obtained by **Abdul Ghaffar *et al.* (2009)**.

Moreover, the concentrations of all trace elements were decreased in all water samples after the exposure to the electromagnetic field for two days , and this may be attributed to the direct effect of the electromagnetic field on chemical characteristics of water and attraction of macromolecules. It is agreed with those of **Kholodov (1974)** and **Alkhazan and Sadddiq (2010**). Also, The decrease can be explained thus, that magnetic force breaks hydrogen bonds between water molecules, so the ions become separated and combine with elements and precipitate **Alkhazan and Sadddiq (2010**). In addition, **Chang and Weng (2008)** showed that the enhanced mobility of the ions under a magnetic field, causes serious damage to the hydrogen bond network in the high Na concentration solution. Conversely, in the low-concentration solution, the structural behavior is dominated by the properties of the water molecules and hence the hydrogen bonding ability is enhanced, as the magnetic field is increased.

Heavy metals in fish tissues:

The constant magnetic field intensifies biological degradation processes by activated sludge of most of the tested organic compounds **Alkhazan and Sadddiq (2010**). It was also confirmed that, the magnetic field's effect on organic compound degradation continues for about 12 hrs after termination of exposure **(Lebkowska, 1991)**.

Muscle tissue:

Regarding Zn levels they were lower than the safe limits (50 mg/kg) recommended by **WHO (1984)** and **USEPA (1986)**. The results obtained in this study are nearly close to those recorded by **Mohamed (2008)**; **Murugan *et al.* (2008)**; **Miclean *et al.* (2009) and Khater (2010),** whereas they are lower than those reported by **Morshdy *et al.* (2007); Uluozlu *et al.* (2007)** and **Al-Bader (2008).** Also, this results found that Zn concentrations were decreased with the electromagnetic exposure. It is in accordance with **Kholodov (1974)** and **Alkhazan and Sadddiq (2010**).

The lead levels in the present study were lower than the permissible limits (2.0 mg/kg) recommended by **WHO (1984)** and **USEPA (1986)**, and are lesser than those recorded by **Al-Bader (2008)**; **Begum *et al*. (2009); Miclean *et al*. (2009).** Moreover, this results found that Pb concentrations were decreased with the electromagnetic exposure. It is in accordance with **[Kholodov (1974)** and **Alkhazan and Sadddiq (2010)**]. Manganese levels in the present study were higher than the permissible limits (2.5 mg/kg) recommended by **WHO (1993)** at the studied species. The results obtained in this study are nearly close to those recorded by **Gabriel *et al*. (2006)**,whereas they are higher than those reported by **Olaif *et al*. (2004); Uluozlu *et al*. (2007)**. However, this results found that Mn concentrations were decreased with the electromagnetic exposure. It is in accordance with **[Kholodov (1974) and Alkhazan and Sadddiq (2010)**].

Aluminium levels in the present study were higher than the permissible limits (0.1 mg/kg) recommended by **ANZECC (2000)** at three studied species. Comparative Al levels were recorded by **Svobodova *et al*. (1993) and Schlotfeldt& Alderman (1995).**this values were decreased with the electromagnetic exposure. It is in accordance with **[Kholodov (1974) and Alkhazan and Sadddiq (2010)**].

Blood, Gills, Kidney and liver tissues:

From the present data, it is obvious that blood, kidney, liver, and gills accumulate higher concentrations of the studied trace elements here than the muscle. The mean of all studied elements of these tissues exceeded the permissible concentrations [(except for Zn and Pb (in gills)]. Blood represented the highest site of concentrations of trace elements followed by kidney, liver, then gills and finally muscle tissue. This may be attributed to the major role of blood, kidney and liver in detoxification and protection from trace elements exposure, both by producing metallothionines (metal binding-proteins) and by acting as storage site for bound metals **(Pratap *et al*., 1989)**. Also, the liver concentrates these elements from the blood circulatory system of the fish.

The present results in agree with **Olaif *et al*. (2004)**; **Yap *et al*. (2005)** and **Velcheva (2006)** that trace elements were significantly higher in fish viscera, including liver tissue and kidney than in the edible muscle tissue. The high trace elements content gills of fish collected from the sources of water can be related to accumulate ion of such trace elements from the water primarily through fish gill where metallothionine enhances that bioaccumulation in gills **(Saeed, 2000)** and its uptake could be controlled by the amount of water passing through the gills.

These results also comply with results obtained by **Mohamed (2008)**; **Murugan *et al*. (2008)**; **Akan *et al*. (2009)**.

In the present study, it is clear that the accumulation of trace elements in muscle, gills, kidney, blood and liver tissue increased with time of exposure and this coincides with **Khater (2010)** who mentioned that the accumulation of trace metals by fish depending on size, age, species and environment of fish. Moreover, it is obvious that the trace elements concentrations were decreased with the electromagnetic exposure. It is in accordance with **[Kholodov (1974) and Alkhazan and Sadddiq (2010)**].

These results may be due to the effect of the electromagnetic field which influence the entire organism, its systems and organs, cells, sub cellular formations, and molecular structures. It was shown that magnetic fields change osmotic processes in muscles, affect the permeability of the cellular membrane, and disturb the hydration ability of tissues in animals. Various mechanisms are suggested as a possible route of the realization of the influence of CMF on biological objects: changes in the orientation of large molecules in strong fields, the inhibiting effect of CMF on the rotational diffusion of large molecules, changes in the angle of the chemical bond in the molecules, changes in the rate of proton tunneling in hydrogen

bonds between nucleotides making up a deoxyribonucleic acid molecule, the influence of CMF on pulsating biocurrents, which may cause mechanical shifts of the sources of biopotentials, and changes in the properties of water in CMF **(Kholodov, 1974)**.

Conclusion

The results of this study concluded that the effects of exposure to extremely low frequency electromagnetic fields depend on the magnetic field intensity, frequency and the period of exposure, and the interference of the applied electromagnetic field with the bioelectrical signals generated during the physiological processes may affect the biological functions causing either enhancement or inhibition of the running processes.

Also, the biological treatment of water using magnetic force has a vital role in treating the polluted water. This is one of the interesting findings in this field of research. This encourages more research in this field. Using magnetism to overcome negative effects of water pollution is considered a potential technology. It should be adapted to suit environmental and climatic conditions so that its use can be maximized.

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