

Effect of *Moringa oleifera* on Hemato-Biochemical and Histological Alterations in Rats after Exposure to High Voltage Electromagnetic Field

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ABSTRACT

Electromagnetic fields (EMFs) have various chemical effects, including deterioration in large molecules in cells and imbalance in ionic equilibrium. The current study investigated the effect of electromagnetic field generated

by high voltage (HV) 5.4 k/v for 2,4h per day with frequency equals 50Hz on body weight, blood indices and some liver enzymes of albino rats after exposing them to the electromagnetic field for 25 days. This work focuses on the therapeutic action of methanolic extract of *Moringa oleifera* (*M. oleifera*) leaves at dose (300 mg/kg B.W) against harmful effects induced by electromagnetic field. Results showed that electromagnetic field exposure caused a significant decrease in an organs weight, RBCs, Hb and CAT. While WBCs, AST, ALT, Total Bilirubin, Urea, Creatinine, Uric Acid and MDA levels were significantly increased under electromagnetic field exposure. Treatment with *M. oleifera* revealed attenuation in these parameters that had been caused in rats exposed to HV. These results were supported by histopathological examination of liver and brain sections

In conclusions, *M. oleifera* leaves extract afforded significant protection against HV induced liver and other organs injury and therefore may have application in the field of drug development.

Key words: Electromagnetic field, *M. oleifera*, hematological parameters, MDA, CAT.

1- INTRODUCTION

Electromagnetic fields (EMFs), a physical field produced by moving electrically charged objects (**Khaki et al., 2011**) and affect the behavior of charged objects in the vicinity of the field are emitted by many natural and man-made sources that play important roles in daily life. It is extending indefinitely throughout space and describes electromagnetic interaction (**Schüz et al., 2009 and Emre et al., 2011**). Mainly four categories of EMFs signals can be identified. They are classified as static, electric and MFs as direct current (DC, 0 Hz), Extremely Low Frequency fields (ELFs), between 1Hz up to 300 Hz. The time-varying EMFs are an example of ELF-EMF. Other technologies produce intermediate frequency (IF) fields with frequencies from 300 Hz to 10 MHz and high frequency (HF)

fields, in the band of the Radio Frequency fields (RF) 10 MHz–300 GHz and of the microwaves (MW) above 3 GHz (**Harrington *et al.* 1997 and ICNIRP 2010**).

Human body exposure to EMF caused various chemical effects, imbalance in ionic equilibrium, biological systems reactions, stress, and oxidative stress in many tissues of the body, physiological and psychological effects on human health. (**Thakare and Utane, 2018**).

Moringa oleifera Lam. is a small size tree with approximately 5 to 10 m height. It is cultivated all over the world due to its multiple utilities. Every part of *Moringa* is used for certain nutritional and/or medicinal propose (**Farooq *et al.*, 2012**) besides being a good source of protein, vitamins (A and C), minerals (Ca and K), oils and fatty acids (**Kamal, 2008; Eshak and Osman, 2013**) which act as natural antioxidants (**Mahmood *et al.*, 2010**).

M. oleifera leaves methanolic extract was rich in phenolic and flavonoid compounds which identified as nutritional and medicinal properties (**Shanmugavel *et al.*, 2018; Dodiya and Amin, 2015**). *M. oleifera* leaves contain polyphenol that potential to have antioxidant (**Fitriana *et al.*, 2016**). It has been reported that, the leaves of *M. oleifera* have various biological activities including anticancer activities, prevention of cardiovascular diseases, Liver disease (**Kumar and Pari, 2003**) antitumor, nervous disorder inflammation digestive disorders, skin disorders, anti-microbial, immunomodulatory and regulation of thyroid status (**Bernett *et al.*, 2003**). All parts of this plant have variously biological activities such as reducing hyperglycemia (**Mbikay, 2012**).

The present study aims to investigate the role of methanolic *M. oleifera* leaves extract against high voltage induced oxidative stress in rats.

2-MATERIALS AND METHODS

2.1 Plant Material:

The leaves of *Moringa oleifera* samples were purchased from the experimental farm Faculty of Agriculture Minia University. The fresh plant leaves were cleaned carefully, washed several times with running tap water and dried for 5 days in the shade. These leaves were ground by monlinex blinder in to fine powder, and kept in the refrigerator at 4°C for preparation crude extracts.

2.2 Preparation of *M. oleifera* extracts:

The extract was prepared by stirring 100 g of plant leaves with 700 ml of a solvent composed of 800 ml methanol and 200 ml distilled water on magnetic stirrer for 24 h at room temperature. The infusion was filtered by a piece of double layer gauze and centrifuged at 3000 r.p.m for 10 min. The residue was re-extracted by the same method. The combined filtrates were evaporated using a rotary evaporator apparatus attached with vacuum pump at 40°C to dryness and the residue kept in a refrigerator. The residue was suspended in distilled water and the dose was 300 mg/kg b.w. (**Khalafalla et al., 2009** with a minor modifications).

2.3 Experimental Animals:

Adult male albino rats of Sprague-Dawley strain, weighting 130±20 g were obtained from the Animal House Faculty of Pharmacy - Al Nahda University – Beni Suef. The rats were housed in plastic cages in air condition room at 25 ± 2 (with a 12 h light/ dark cycle). A commercial balanced diet and tap water were provided *ad libitum* for two weeks before starting the experiment. All experimental procedures were conducted according to the ethical standards approved by the Institutional Animal Ethics Committee guidelines for animal care and use, Minia University, Egypt.

2.4 Experimental Design:

Forty eight healthy rats were randomized assigned into 6 groups (8 rats each).

- (1) Control group: Rats were received distilled water.
- (2) Moringa group: Rats were received Moringa extract 300 mg / kg (**Mansour 2013**).
- (3) H.V 2 h group: Rats were exposed to high voltage for two hours.
- (4) H.V 4 h group: Rats were exposed to high voltage for four hours.
- (5) Moringa + H.V 2 h group; Rats were received extract 300 mg / kg for 5 days before and exposed to H.V 2 h concurrently with Moringa for 25 days.
- (6) Moringa + H.V 4 h group: Rats were received Moringa extract 300 mg / kg for 5 days before and exposed to H.V 4 h concurrently with Moringa for 25 days.

2.5 Electromagnetic field exposure:

The experiment is done in Electrical Department. Faculty of Engineering, Assiut University. The used system consists of two parts; first part is transformer that used to convert the current from low voltage to high voltage. The second part is exposure area consisting of two plates. The upper plate is made from copper and the other from Aluminium. These plates connected to the transformer and the cage of rats put in between. The distance between two plates is 1.8m. The cage was made from plastic, dimensions 46 cm long and 16 cm wide and 20 cm high. The groups were exposed to 50 Hz, 5.4Kv electric field for a period of 25 days (2h and 4h/day, 5 days/week) and the rats were weighed weekly.

2.6 Sampling

2.6.1 Blood samples

At the end of 30 days, rats were fasted overnight and anesthetized to collect the blood samples from the retro-orbital plexus (**Schermer, 1967**).

Suitable volumes of fresh blood were immediately taken in heparinized tube for hematological examinations. The other parts of blood samples were allowed to coagulate at room temperature, and then centrifuged at 4°C the clear non-haemolysed sera were separated and stored at -20°C till used in biochemical analysis.

2.6.2 Tissues samples

Rats were sacrificed and the organs, liver, kidney, lung, spleen, tests, heart and brain were excised, wiped with filter paper and weight. Small parts of liver and brain were fixed in 10% formalin solution for histopathological examinations. Other parts of liver tissue were homogenized and used for measuring CAT and MDA concentrations.

2.7 Hematological evaluation

Blood was taken from the retro-orbital plexus in heparinized tube for determination red blood cells (RBCs) and white blood cells (WBCs) counts, haemoglobin concentration (Hb), packed cell volume (PCV), mean cell volume (MCV), mean corpuscular haemoglobin (MCH), mean corpuscular hemoglobin concentration (MCHC) using Animal Blood Counter (Genius-KT-6400).

2.8 Biochemical assays

Serum enzyme activity of aspartate aminotransferase (AST; E.C.2.6.1.1.) and alanine aminotransferase (ALT; E.C.2.6.1.2.) activities were measured according to the method described by **Reitman and Frankel, (1957)**. Total protein, albumin, urea, creatinine and uric acid level were determined according to the methods described by **Gornall *et al.*, (1949)**; **Doumas *et al.*, (1971)**; **Fawcett and Soctt, (1960)**; **Bartles *et al.*, (1972)** **Barham and Trinder, (1972)**. Globulin was determined by difference between total protein and albumin. Total and direct bilirubin were determined calorimetrically (**Walters and Gerarde, 1970**). Total lipids (TL), Triglycerides (TG), Cholesterol(TC), HDL-Cholesterol and LDL-Cholesterol were determined according to the methods described by

(Zollner and Kirsch, (1962); Fassati and Prencipe (1982); Allain, *et al.*, (1974); Burstein, *et al.*, (1970) and Wieland and Seidel (1983), using enzymatic colorimetric procedures Kits from Bio- Diagnostic Co., Egypt.

2.9 Measuring of liver CAT and MDA

Half gram of liver tissue from each animal was homogenized in 5 ml of 100 mM phosphate buffer, pH 7.4 on ice using Universal laboratory Aid homogenizer, homogenates were then centrifuged at 3000 rpm for 15 min at 4°C. The resulting supernatants were collected and preserved at -20 °C until measuring of catalase activity (CAT) and Malondialdehyde (MDA) concentration according to methods described by Aebi (1984) and Ohkawa *et al.*, (1979).

2. 10 Histopathological examination:

Liver specimens were fixed in 10% formalin solution. After 24 h of fixation followed by embedding in a paraffin block, it was cut into sections of 5µm thickness and stained with hematoxylin -eosin (H&E) for routine histopathological examination according to Bancroft *et al.* (1996).

2. 11. Statistical analysis:

The results obtained in the present study were evaluated by One Way ANOVA test. The results were expressed as mean \pm standard error and values of $P < 0.05$ were considered statistically significant (Snedecor and Cochran, 1986).

3-Results and Discussion

3. 1. Effect on body weight and feed efficiency of rats

Results in Table (1) showed that there were no significant differences in *M. oleifera* extract treated groups compared to normal control in body weight gain and feed efficiency ratio for 30 days. Our results were in agreement with that for El-bakry *et al.* (2016).

Table 1 Effect of *M. oleifera* (300 mg / kg b. w) on body weight gain, daily feed intake and feed efficiency ratio on rats exposed to high volt 2 and h for 25days.

Groups	Final weight (g)	Body weight gain (g)	Daily body weight gain.(g)	Daily feed intake (g)	Feed efficiency ratio (%)
Control	239.30±13.41	87.81±14.01	3.53±0.61	15	23.55±3.81
M	236.43±9.30	84.41±6.10	3.41±0.24	15	22.61±1.62
HV 2h	235.43±10.10	62.98 ^a ±6.41	2.65 ^a ±0.20	15	16.87 ^a ±1.60
HV 4h	227.31 ^a ±6.54	60.58 ^a ±8.21	2.43 ^a ±0.33	15	16.15 ^a ±2.21
M & HV 2h	242.50 ^{ab} ±10.97	70.82 ^{ab} ±6.24	2.84 ^a ±0.24	15	18.92 ^a ±1.70
M & HV 4h	237.40±3.13	63.45 ^{ab} ±4.10	2.54 ^a ±0.20	15	16.95 ^a ±1.11

Data represent the mean ±S.E. of observations from eight rats. a and b significantly different from control and high volt exposed groups at $P < 0.05$ respectively .

The body weight gain and feed efficiency ratio were significantly ($P < 0.05$) decreased in rats exposed to high voltage for 2, 4 h about 28 and 31% respectively compared to the control group. Meanwhile pretreated groups with methanolic *M. oleifera* extracts reduced significantly ($P < 0.05$) the reduction of body weight gain and caused slight improvement in the feed efficiency ratio compared with exposed group to high voltage for 2, 4 h. *M. oleifera* contains phenolic compounds as kaempferol, rhamnetin, quercetin, chlorogenic acid, rutin, apigenin which consider antioxidant activity (Karthivashan *et al.*, 2013; Samani *et al.*, 2015).

3. 2.Changes in relative weight of some organs:

Results in Table (2) reveal that the relative weight of liver, kidney, testis and heart were decreased but not significantly ($P < 0.05$) meanwhile, lung, spleen and brain increased but not significantly in all groups compared to control group.

Table 2 Effect of *M. oleifera* (300 mg / kg b. w) extract on relative organs of rats exposed to high volt 2 and h for 25days.

Groups	liver%	kidney%	Lung%	Spleen%	Testis%	Heart%	Brain%
Control	10.87±1.16	1.90±0.16	1.53±0.11	1.12±0.08	2.87±0.32	1.07±0.09	1.71±0.05
M	10.00±0.38	1.98±0.09	1.52±0.04	1.84±0.17	2.83±0.19	0.92±0.02	1.64±0.04
HV 2h	10.71±0.25	1.84±0.06	1.79±0.08	1.43±0.04	2.78±0.10	1.14±0.03	1.76±0.03
HV4h	10.80±0.34	1.82±0.06	1.75±0.04	1.44±0.08	2.75±0.08	1.04±0.04	1.74±0.02
M & HV 2h	10.18±0.45	1.78±0.04	1.92±0.09	1.43±0.09	2.52±0.31	1.01±0.05	1.71±0.03
M & HV 4h	10.79±0.49	1.79±0.03	1.79±0.03	1.54±0.07	2.41±0.20	1.01±0.06	1.78±0.03

Data represent the mean ±S.E. of observations from eight rats. a and b significantly different from control and high volt groups at $P < 0.05$ respectively .

Our results were similar to the obtained data by Saalu *et al.*, (2012) who observed that liver weights, liver weight/body weight ratio and volumes were lower but not significant comparing with control and moringa alone.

3. 3. Effect of *M. oleifera* extract with exposure high voltage on hematological parameters

The hematological studies of the animals administrated with them ethanolic *M. oleifera* extract for 30 days showed no significant differences in the RBCs, Hb, and WBCs levels compared to the control (Table 3).

The RBCs was significantly ($P < 0.05$) decreased in exposed groups to high voltage for 2,4 h about 12 and 6% and Hb about 15 and 11% respectively comparing with control group. For the meanwhile, pretreated groups with methanolic *M. oleifera* extract improved significantly ($P < 0.05$) this reduction and restored it to nearly normal control.

Table3. Effect of *M. oleifera* (300 mg / kg b. w) extract on hematological parameters in blood rats exposed to high volt 2 and 4 hours for 25days.

Groups	RBCs ($10^6/\text{mm}^3$)	HB (mg/dl)	PCV (mg/dl)	WBCs ($10^3/\text{mm}^3$)	MCV $\times 10^{-15}$	MCH	MCHC
Control	6.34±0.41	13.88±0.64	0.37±0.37	12.33±1.66	47.80±0.79	21.15±0.56	44.30±0.58
M	6.36±0.55	13.85±0.82	0.36±0.36	11.85±1.91	48.50±0.17	21.20±0.38	44.25±0.36
HV2h	5.56 ^a ±0.69	11.80 ^a ±0.89	0.28 ^a ±0.15	15.00 ^a ±1.01	49.30 ^a ±0.77	21.50±0.34	44.40±1.18
HV4h	5.94 ^a ±0.48	12.40±0.78	0.26 ^a ±0.06	19.03 ^a ±6.27	49.70 ^a ±0.39	21.90±0.41	44.50±0.49
M& HV2h	5.85±2.31	12.60±2.46	0.35 ^b ±0.05	12.40 ^b ±2.23	47.20 ^b ±1.29	20.80±1.01	44.90±2.27
M& HV4h	6.30±0.78	13.70±1.11	0.32 ^b ±0.04	15.20 ^{ab} ±3.01	47.60 ^b ±0.54	21.05±0.49	44.40±0.62

Data represent the mean ±S.E. of observations from eight rats. a and b significantly different from control and high volt groups at $P < 0.05$ respectively .

Packed cell volume (PCV) was significantly ($P < 0.05$) decreased in the exposed group to high voltage for 2,4 h about 24 and 30% respectively compared to the control group. Meanwhile, pretreated groups with methanolic *M. oleifera* extract improved significantly and returned to normal control.

The WBCs was significantly ($P < 0.05$) increased in the exposed groups to high voltage for 2,4 h about 22 and 54% respectively compared to the control group. Whereas, pretreated groups with methanolic *M. oleifera* extract improved significantly ($P < 0.05$) this increase of WBCs about 17% compared to exposed group to high voltage for 2 h, Meanwhile this improved was 20% comparing with exposed group to high voltage for 4 h.

The increasing number of white blood cells attributed to the variations of states and properties of the protein molecules such as the hemoglobin in blood (Xiao-Feng and Gun, 2017). Also, the depletion in the values of hematological parameters following EMF radiation exposure may be attributed to direct damage caused by radiation and due to overproduction of Reactive Oxygen Species by microwave radiation interaction.(Eid *et al.*, 2015). It has also been reported that the production of free radicals cause hemolysis (Aweda *et al.*, 2004).

3. 4. Effect of *M. oleifera* extract with exposure high voltage on liver function

The enzymes of ALT and AST are critical for biological processes that occur within the body of a living organism and are one of the signs of the health. Data in Table (4) showed that there were no significant differences in *M. oleifera* extract treated groups compared to normal control in AST and ALT activity for 30 days.

Table4. Effect of *M. oleifera* (300 mg / kg b. w) extract on liver functions in rats exposed to high volt2 and 4 h for 25days.

Groups	AST (U/ml)	ALT (U/ml)	T. Bilirubin (mg/dl)	D. Bilirubin (mg/dl)	Ind.Bilirubin (mg/dl)
Control	40.10 ±0.03	45.10±0.00	1.02±0.01	0.30±0.01	0.73±0.01
M	40.23 ±0.04	45.10±0.00	1.03±0.01	0.30±0.01	0.72±0.00
HV 2h	60.13 ^a ±0.05	58.84 ^a ±0.04	1.45 ^a ±0.01	0.59 ^a ±0.01	0.86 ^a ±0.01
HV 4h	64.56 ^a ±0.03	61.44 ^a ±0.05	1.68 ^a ±0.01	0.72 ^a ±0.01	0.96 ^a ±0.01
M &HV 2h	43.76 ^{ab} ±0.0	54.81 ^{ab} ±0.05	1.16 ^{ab} ±0.01	0.46 ^{ab} ±0.01	0.70 ^{ab} ±0.00
M &HV 4h	44.63 ^{ab} ±0.07	56.26 ^{ab} ±0.06	1.23 ^{ab} ±0.01	0.52 ^{ab} ±0.01	0.72 ^b ±0.01

Data represent the mean ±S.E. of observations from eight rats. a and b significantly different from control and high volt groups at $P < 0.05$ respectively.

Meanwhile, exposure to high voltage for 2, 4 h increased significantly ($P < 0.05$) the serum AST activity about 50 and 61% respectively whereas ALT activity 30 and 36% comparing with control group. Whereas the exposure groups to high voltage for 2, 4 h concurrently with *M. oleifera* significantly ($P < 0.05$) reduced the serum levels of these enzymes about 31% comparing with groups exposed to high voltage for 2, 4 h. All liver function tests are influenced as a result of exposure to extremely low frequency magnetic fields and mostly affected with a percentage $> 90\%$ (Ibrahim *et al.*, 2008).

At the same Table (4) results showed that there were no significant differences in *M. oleifera* extract treated groups compared to normal control in T.B, D. B and I.B activity for 30 days. Meanwhile, exposure to high voltage for 2, 4 h increased significantly ($P < 0.05$) the serum T.B activity about 42 and 65% respectively whereas D. B levels 97 and 140% while I. B levels 18 and 32% comparing with control group. The elevation in the levels of serum bilirubin may result from the damaged cells which leak into circulation after exposure to magnetic field (Novikov *et al.*, 1999).

Exposure groups to high voltage for 2, 4 h concurrently with *M. oleifera* significantly ($P < 0.05$) reduced the serum levels of T.B and D.B about 37% and 27% comparing with groups exposed to high voltage for 2, 4 h respectively. While the serum levels of I. B was restored to nearly control.

M. oleifera extract showed significant improvements in liver function tests in animals exposed to gamma irradiation (Eshak and Osman, 2013). These may be attributed to the stabilizing ability of the cell membrane preventing enzymes leakages (Pari and Karthikesan, 2007) *M. oleifera*, is considered one of the herbal drugs contains a variety of chemical constituents like phenols, coumarin's, lignin's, essential oil, monoterpenes, carotenoids, glycosides, flavonoids, organic acids, lipids, alkaloids and xanthene's (Gupta and Misra, 2006).

3. 5. Effect of *M. oleifera* extract with exposure high voltage on kidney function parameters

Results in Table (5) showed that there were no significant differences in *M. oleifera* extract treated groups compared to normal control in urea, uric acid and creatinine activity after 30 days. Meanwhile exposure to high voltage for 2,4 h increased significantly ($p < 0.05$) the serum urea levels about 24 and 40% respectively whereas creatinine increased about 82 and 108%. Uric acid levels increased about 74 and 82 % comparing with control group. However the serum levels of the urea, uric acid and creatinine revealed a significant ($p < 0.05$) decrease in a treatment with *M. oleifera* extract. *M. oleifera* concurrently with the exposure to high voltage for 2 h showed a slight decreased in urea concentration. While after 4h urea was decreased about 14% comparing with group exposed to high voltage for 4 h.

Table5. Effect of *M. oleifera* (300 mg / kg b. w) extract on Kidney function in rats exposed to high volt 2 and 4 hours for 25days.

Groups	Urea (mg/dl)	Creatinine (mg/dl)	Uric Acid (mg/dl)
Control	28.41±0.02	2.64±0.05	4.59±0.02
M	28.40±0.05	2.87±0.03	4.64±0.01
H.V2h	35.31 ^a ±0.15	4.81 ^a ±0.02	7.98 ^a ±0.02
H.V4h	39.63 ^a ±0.06	5.49 ^a ±0.03	8.33 ^a ±0.01
H.V2h&M	33.47 ^{ab} ±0.07	3.25 ^{ab} ±0.05	6.43 ^{ab} ±0.01
H.V4h&M	33.91 ^{ab} ±0.03	3.45 ^{ab} ±0.05	6.73 ^{ab} ±0.08

Data represent the mean ±S.E. of observations from eight rats. a and b significantly different from control and high volt groups at $P < 0.05$ respectively

M. oleifera concurrently with the exposure to high voltage for 2,4h showed significantly ($p < 0.05$) decrease of creatinine about 36% and uric acid about 19% comparing with groups exposed to high voltage 2,4 h.

Irradiation may cause breaking of DNA molecules and destruction of their bases (the purines) which may be catabolized into uric acid (Ganong, 1999). Creatinine is formed largely in muscles and occurs freely in blood plasma and urine.

M. oleifera has been shown to possess diuretic effect (Mbikay, 2012 and Kumar *et al.*, 2010) and this may have contributed for protecting against homeostatic imbalance imposed by HV exposure. *M. oleifera* may

be induced cell membrane fixation ability and thus inhibit enzyme leakage and restore cell integrity (Pari and Karthikesan, 2007). Also attributed to the presence of certain antioxidants such as quercetin and amphetamine (Selvakumar and Natarajan, 2008), Vit A and C (Toppo *et al.*, 2015).

3. 6. Effect of *M. oleifera* extract with exposure high voltage on Total Protein and Albumin parameters

Results in Table (6) showed that there were no significant differences in *M. oleifera* extract treated groups compared to normal control in protein and albumin levels after 30 days. Meanwhile exposure to high voltage for 2, 4 h increased significantly ($p < 0.05$) the serum protein level about 39.5 and 43.1%, respectively whereas albumin levels 58 and 75 % comparing with control group. While *M. oleifera* concurrently with the exposure to high voltage for 2,4h significantly ($p < 0.05$) decreased the serum level of protein about 20 % and albumin about 22% comparing with high voltage for 2,4 h groups, respectively.

Table 6. Effect of *M. oleifera* (300 mg / kg b. w) extracts on total protein and albumin in rats exposed to high volt 2 and 4 hours for 25days.

Groups	Protein (mg/dl)	Albumin (mg/dl)	Globulin (g/dl)	Albumin /globulin
Control	7.70±0.01	4.12±0.00	3.57	1.15
M	7.81±0.01	4.30±0.00	3.56	1.19
HV 2h	10.74 ^a ±0.01	6.51 ^a ±0.00	4.28	1.51
HV 4h	11.02 ^a ±0.01	7.21 ^a ±0.00	3.84	1.87
M & HV 2h	8.61 ^{ab} ±0.01	5.37 ^{ab} ±0.00	3.24	1.66
M & HV 4h	8.85 ^{ab} ±0.01	5.60 ^{ab} ±0.00	3.30	1.68

Data represent the mean ±S.E. of observations from eight rats. a and b significantly different from control and high volt groups at $P < 0.05$ respectively .

The formation of ROS may increase the dietary oxidized fats and may cause an increased damage of proteins in the liver by enhancing lipid per oxidation of the cell membrane and increasing the generation of ROS which can lead to calcium homeostasis disturbances, increase membrane fluidity and cell death (Hassanen, 2015).

M. oleifera extract showed improvements in serum albumin and total proteins levels due to an early improvement in the cellular membrane integrity of the hepatic cell which is a clear manifestation of anti-hepatotoxic effect of its administration. The protective mechanisms of *M. oleifera* leaves extract may follow an antioxidant mediated mechanism (El-bakry *et al.*, 2016 and Onah *et al.* 2016). *M. oleifera* is considered as a high delivery source of protein, β -carotene, vitamins A, B, C, E, riboflavin, nicotinic acid, folic acid and pyridoxine, amino acids, minerals and various phenolic compounds (Anwar *et al.*, 2007; Khalafalla *et al.*, 2010 and Fakurazi *et al.*, 2012).

3. 7. Effect of *M. oleifera* extract with exposure high voltage on lipid profile parameters

Results in Table (7) showed that there were no significant differences in *M. oleifera* extract treated groups compared to normal control in TL, TC, TG, HDL, LDL and VLDL activity after 30 days. Meanwhile exposure to high voltage for 2, 4 h increased significantly ($p < 0.05$) the serum TL activity about 98 and 114% respectively whereas TC, TG, LDL and VLDL levels about 19, 48, 105 and 48% in high voltage for 2 h while 22, 63, 136 and 63 % in high voltage for 4 h comparing with control group. But on the contrary exposure to high voltage for 2, 4 h reduced significantly ($p < 0.05$) the serum HDL activity about 48 and 67% respectively compared with control group. Meanwhile *M. oleifera* concurrently with the exposure to high voltage for 2, 4h significantly ($p < 0.05$) decreased TL about 9 and 12% and showed a slight improvement in TC level comparing with high voltage for 2, 4h exposure group respectively.

Table 7. Effect of *M. oleifera* (300 mg / kg b. w) extract on lipid profile in rats exposed to high volt 2 and 4 hours for 25days.

Groups	TL (mg/dl)	TC mg/dl	TG mg/dl	HDL mg/dl	LDL mg/dl	VLDL mg/dl
Control	566.67±2.27	197.73±0.18	150.00±1.02	110.89±0.30	86.93±0.34	30.00±0.20
M	577.21±2.26	198.37±0.31	158.13 ^a ±3.13	110.96±0.22	88.41±0.57	31.75±0.52
HV2h	1122.81 ^a ±1.43	235.70 ^a ±0.15	221.25 ^a ±1.61	57.24 ^a ±0.48	178.31 ^a ±0.29	44.25 ^a ±0.32
HV4h	1214.04 ^a ±1.43	241.62 ^a ±0.31	244.38 ^a ±1.19	37.16 ^a ±0.59	205.22 ^a ±0.34	48.88 ^a ±0.24
M&HV2h	1026.32 ^{ab} ±4.18	228.43 ^{ab} ±0.15	187.50 ^{ab} ±1.02	74.753 ^{ab} ±0.49	155.54 ^{ab} ±0.34	37.50 ^{ab} ±0.20
M&HV4h	1066.67 ^{ab} ±19.95	232.51 ^{ab} ±0.09	202.50 ^{ab} ±1.02	60.31 ^{ab} ±0.55	171.21 ^{ab} ±0.57	40.50 ^{ab} ±0.20

Data represent the mean \pm S.E. of observations from eight rats. a and b significantly different from control and high volt groups at $P < 0.05$ respectively.

At the same time *M. oleifera* concurrently with the exposure to high voltage for 2,4 h caused a significant ($p < 0.05$) decreased in TG and VLDL about 15 and 17% comparing with high voltage for 2,4 h group respectively. While LDL was decreased about 17% comparing with groups exposed to high voltage for 2,4 h respectively.

On the contrary with *M. oleifera* concurrently with the exposure to high voltage for 2,4h significantly ($p < 0.05$) increased HDL about 31 and 62% comparing with high voltage for 2,4h group respectively.

M. oleifera leaves extract had lipid lowering effect particularly of TG and VLDL, is an encouraging one. Lipids have been implicated in the pathogenesis of inflammatory diseases, such as rheumatoid arthritis and cardiovascular diseases (Ara *et al.*, 2008).

3. 8. Effect of *M. oleifera* extract with exposure high voltage on oxidative stress parameters

Results in Table (8) showed that there were no significant differences in *M. oleifera* extract treated group compared to normal control in catalase and MDA levels after 30 days. Meanwhile, the exposure to high voltage for 2,4h increased significantly ($p < 0.05$) the MDA level about 36.8 and 42.4% respectively whereas decreased significantly ($p < 0.05$) the catalase activity about 20.2 and 30.4 % compared with control group. *M. oleifera* concurrently with the exposure to high voltage for 2,4h caused a significant reduction in MDA about 18 and 14% and significant ($p < 0.05$) elevation in catalase level about 31 and 5% comparing with groups exposed to high voltage for 2,4 h respectively

Table 8. Effect *M. oleifera* (300 mg / kg b. w) extract on hepatic oxidative stress parameters in rats exposed to high volt 2 and 4 hours for 25days.

Groups	CAT (U/L)	MDA(nmole/g)
Control	2.63±0.01	4.95±0.04
M	2.71±0.01	4.73±0.04
HV 2h	2.10 ^a ±0.01	6.77 ^a ±0.03
HV 4h	1.83 ^a ±0.01	7.05 ^a ±0.03
M&HV2h	2.38 ^{ab} ±0.02	5.54 ^{ab} ±0.03
M& HV4h	2.20 ^{ab} ±0.00	6.08 ^{ab} ±0.03

Data represent the mean ±S.E. of observations from eight rats. a and b significantly different from control and high volt groups at $P < 0.05$ respectively .

Changes in MDA levels indicate increased ROS production occurring during the exposure period that may reflect the pathological process of EMF exposure (Fehmi *et al.*, 2005). Many studies have shown that EMF exposure is capable of causing the substantial oxidative damage to the body (Sharma *et al.*, 2017; Sharma *et al.* 2014; Aydin and Akar *et al.*, 2011). Electromagnetic field may be harmful by accelerating the loss of hepatocyte plasma membrane integrity (Đinđić *et al.*, 2010).

3. 9.Histological examination

Histological changes were screened to support the tested biochemical markers of organs injury. The histopathological results showed that *M. oleifera* alone were found to be safe and did not induce any histopathological changes in organs. On the other hand, the pretreated groups with *M. oleifera* effective (Table 9 and Fig1). It is noticed that these sections taken from liver of rats exposed to high voltage for 2, 4 h showed several alterations such as kupffer cells activation, congestion of central vein and sinusoids, hydropic degeneration of hepatocytes and fibroplasia in the portal triad with only appearance focal hepatic necrosis associated with inflammatory cells infiltration in group exposed to high voltage for 4 h.

Table 9.Histopathological notes on liver tissue of rats exposed to high voltage for 2, 4 h plus *M. oleifera* .

Portal fibroplasia	Focal hepatic necrosis associated with inflammatory infiltration	Vacuolation or hydropic degeneration of hepatocytes	Congestion of central vein and sinusoids	Kupffer cells activation	Histopathological lesion
-	-	-	-	-	Control
-	-	-	-	-	M
+	-	++	++	+	H.V2h
++	-	++	++	++	H.V4h
-	-	-	-	+	M&H.V2h
-/++	-/+	-	-	++	M&H.V4h

(-) no, (+) mild, (++) moderate and (+++) severe histopathological changes

Our results agreement with **Hashem and El-Sharkawy (2009)** who found that extremely low-frequency electromagnetic fields (ELF-EMF) caused congestion of hepatic blood vessels and sinusoids, mild lymphocytic infiltration with mild fibroblast, proliferative biliary epithelium and round cell infiltration in liver of mice hepatic cells.

However, pretreatment with *M. oleifera* improved the histopathology of hepatic cells, with only mild kupffer cells activation in group exposed to high voltage for 4 h and a slight for 2h.

Sections taken from brain of rats exposed to high voltage for 2, 4 h showed sever necrosis, pknosis and atrophy of neurons (Fig. 2) and neuronophagia associated with mild congestion of cerebral blood vessel, focal gliosis and cellular oedema. However appearance a moderate necrosis of neurons and focal gliosis in rats exposed to high voltage for 4 h. plus *M. oleifera* and high voltage for 2 h. plus *M. oleifera* showed a moderate necrosis of neurons only.

Histopathological examination of liver:

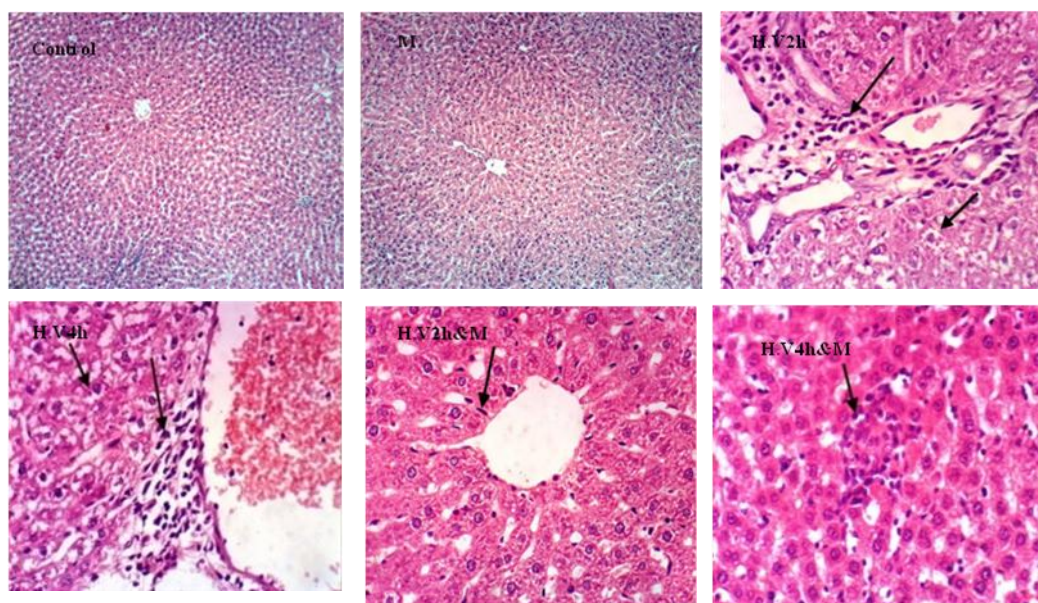


Fig. 1 A photomicrograph sections of liver groups of control, administrated with methanolic *M. oleifera* extract, exposed to HV 2h, exposed to HV 4h, administrated with methanolic *M. oleifera* extract concurrently with HV 2h exposure and administrated with methanolic *M. oleifera* extract concurrently with HV 4h exposure. (H&E, x 100).

Our data agree with **Eser et al., (2013)** who showed exposure to electromagnetic waves (900,1800 and 2450 MHz) caused severe degenerative changes, shrunken cytoplasm, and extensively dark pyknotic nuclei in neurons of the frontal cortex and brain stem tissues in rat.

Table 10. Histopathological notes on brain tissue of rats exposed to high voltage for 2, 4 h plus *M. oleifera*.

Congestion	Cellular oedema	Gliosis	Neuronophagia	Necrosis of neurons	Histopathological lesion
-	-	-	-	-	control
-	-	-	-	-	M
++	+	++	+	+++	H.V2h
++	++	++	+++	+++	H.V4h
-	-	-	+	++	M&H.V2h
+	-	++	+	++	M&H.V4h

(-) no, (+) mild, (++) moderate and (+++) severe histopathological changes

Histopathological examination of brain:

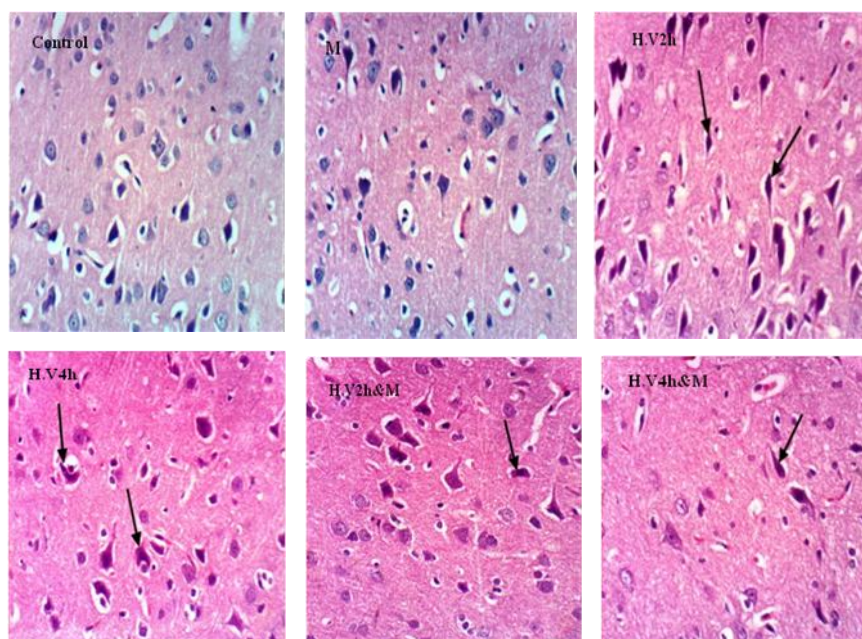


Fig. 2A photomicrograph sections of brain groups of control, administrated with methanolic *M. oleifera* extract, exposed to HV 2h, exposed to HV 4h, administrated with methanolic *M. oleifera* extract concurrently with HV 2h exposure and administrated with methanolic *M. oleifera* extract concurrently with HV 4h exposure. (H&E, x 100).

Conclusion:

The present study concluded that treatment rats with *M. oleifera* leaves alone did not show any significant changes in all parameters when compared with control group. While, pretreated rats with methanolic *M. oleifera* extract able to significantly reduced the oxidative stress induced by high voltage in rats.

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تأثير نبات المورينجا أوليفيرا على التغيرات الدموية والكيميائية والهستولوجية في الجرذان بعد التعرض للمجال الكهرومغناطيسي عالي الجهد

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المستخلص

للمجالات الكهرومغناطيسية (EMFs) تأثيرات كيميائية مختلفة ، حيث إنها تسبب تدهور للجزيئات الكبيرة في الخلايا بالإضافة إلى خلل في التوازن الأيوني. استقصت الدراسة الحالية تأثير المجال الكهرومغناطيسي الناتج عن الجهد العالي (HV) 5.4 كيلو فولت / ساعة لمدة ساعتين و 4 ساعات في اليوم مع تردد يساوي 50 هرتز على وزن الجسم ومؤشرات الدم وبعض أنزيمات الكبد فى جرذان الألبينو بعد تعريضها للمجال الكهرومغناطيسي لمدة 25 يوما. يركز هذا العمل على العمل العلاجي للمستخلص الميثانولي لأوراق المورينجا أوليفيرا (*M.oleifera*) بجرعة (300 مج / كجم من وزن الجسم) ضد الآثار الضارة الناجمة عن المجال الكهرومغناطيسي. أظهرت النتائج أن التعرض للمجال الكهرومغناطيسي تسبب في انخفاض كبير في وزن الأعضاء ، كرات الدم الحمراء ، HB و CAT. بينما تم زيادة مستويات WBCs و AST و ALT و Total الكهرومغناطيسي. كشفت المعاملة باستخدام *M. oleifera* إلى تخفيف هذه المعايير التي حدثت في الفئران بعد تعرضها لـ HV. وأيد هذه النتائج الفحص الهستوباثولوجي لقطاعات الكبد والمخ.

الخلاصة : إن مستخلص أوراق *M. oleifera* يوفر حماية كبيرة ضد إصابات الكبد وغيرها من إصابة الأعضاء وبالتالي قد يكون له تطبيق في مجال تطوير الدواء.