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Effects of Low-frequency Electromagnetic Waves on the Spleen, Liver, and Kidney Weight and Therapeutic Role of Vitamin C in Mice

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ABSTRACT

Electronic devices generate electromagnetic fields, and the recent increase in their use has urged researchers to investigate the effects of electromagnetic fields on human health. The present study aimed to evaluate the effects of extremely low-frequency electromagnetic fields on the weight of spleen, liver, and kidneys and the therapeutic role of vitamin C in mice. This experimental study was conducted on 24 adult male mice (BALB/c), which were divided into three groups. The control group included eight mice, which were kept in normal conditions. Another included eight mice with exposure to an electromagnetic field (ELF) with low frequencies (50Hz and 4Mt) for 15 days, and the third group (n=8) had ELF exposure (50Hz and 4Mt) and received vitamin C for 15 days, with vitamin C injected intraperitoneally seven times. After 15 days, the mice were weighed, and the collected samples were dissected. The spleen, liver, and kidneys of the animals were removed at the final stage for the measurements. Data analysis was performed using one-way analysis of variance (ANOVA). According to the results, the ELFs caused a significant reduction in the weight of the spleen and liver of the animals. Furthermore, the appropriate dose of vitamin C could decrease the damage caused by the ELF frequency of 50 Hz.

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Introduction

In recent decades, the increased use of electromagnetic and mobile devices at home, workplaces, and hospitals has urged researchers to assess their possible effects on human health [1, 2]. In daily life, the applied devices most commonly have the frequency of 50-60 Hz [3-5] and current of 6-10 amperes. Depending on the current intensity and distance from the device, the produced magnetic field of these devices has been estimated at 0.1-8 [6]. Based on wave classification, poor magnetic fields have infrared, radio, and microwave wavelengths. In addition, they have medium or short heat-producing waves [7], which are resulted from the presence or movement of charges in the conductor medium [8]. These waves are a linear function of a potential difference, as well as a function of the current.

Current frequency also influences the production of electromagnetic waves [9]. In a study in this regard, Nakamura et al. (2003) investigated the effects of mobile phone waves with the frequency of 915 MHz on 66 pregnant rats, observing that exposure to these waves with the special absorption of 0.6 milliWt/cm^2 and 0.14 Wt/kg for 90 minutes changed placental circulation and decreased the volumetric flow rate [10]. On the other hand, Stankiexcz et al. (2010) evaluated the effects of electromagnetic waves on neutrophils and monocytes, reporting that the waves could decrease cell phagocytosis [11]. Some studies have demonstrated that leakage microwave ovens could alter the steroid levels in mice [12, 13]. Evidence also suggests that diffused microwaves could reduce growth and increase T_4 and cortisol levels in mice [14].

Vitamin C is a widely used dietary compound and antioxidant [15], which functions based on physiological and metabolic reactions against the damage induced by radiation exposure through free radical scavenging and their conversion into the non-free radical form [16, 17]. Regardless of the type of radiation exposure, the induced changes are caused via

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the elevation of oxidative stress. In addition, the imbalance between the production and neutralization free radicals (especially reactive oxygen species) could lead to biochemical changes [18, 19]. Antioxidants such as vitamin C have been shown to diminish these damages and exert protective effects.
The present study aimed to investigate the effects of extremely low-frequency electromagnetic fields on the weight of the spleen, liver, and kidneys and role of vitamin C in mice.

Materials and Methods

This experiment was conducted at the Research Electrophysiology Laboratory of the Department of Biochemistry and Biophysics of Azad University, Mashhad Branch in Mashhad, Iran. To evaluate the effects of electromagnetic waves with the frequency of 50 Hz on the biomass, liver/kidney biometry, and therapeutic effects of vitamin C on mature mice, BALB/c mice aged 2.5-3 months with the mean weight of 20-25 grams were selected for the study. The mature mice were maintained in an animal room with 60-70% humidity at the temperature of 1±23°C within a 12-hour light/dark cycle overnight. During the 15-day study period, the animals were divided into three groups, including control (eight mice kept in an animal room with normal conditions), exposure to electromagnetic waves (eight mice exposed to electromagnetic waves of 50 Hz/4 mT for 15 days four hours per day; total: 12-16 hours), and exposure to electromagnetic waves and receiving vitamin C (eight mice exposed to electromagnetic waves of 50 Hz/4 mT for 15 days and treated with 0.5 cc of intra-percutaneous vitamin C seven times). The injection was performed three times per week. The lighting of the room was adjusted alternatively by an automatic electrical timer, and the temperature was controlled by radiators and a cooler in winter and summer, respectively. The cages of the animals were cleaned every other day, and wood chips were used for the smoothness of the cages. In addition, the mice feed was prepared in special pellets (Khorasan Javaneh Dam Co.), and water was delivered by flasks. In this study, the mice were purchased from Razi Vaccine and Serum Research Institute of Mashhad, Iran. For adaptation to the new environment and eliminating the stress of changing the environment of the animals, they were kept in an animal room one week prior to the experiments.
The electromagnetic wave-producing machine had the diameter of 35 centimeters and length of 60 centimeters in the form of a PVC tube equipped with 1,900 copper coils, which were coiled three times around the tube. The machine could produce the electromagnetic field of 0.5-4 mT/25-100 Hz. In order to evaluate the therapeutic role of vitamin C, commercial 250 Osveh vitamin C tablets (chewable scored tablets) were used. In order to inject the tablets, they were dissolved in 50 cc of physiological serum. Intraperitoneal vitamin C was injected using a disposable syringe. During the experiments, the mice were weighed three times weekly using a scale at the resolution of 0.01, and the data were recorded. To ensure the accurate weighing of the animals, the process was performed in triplicate. At the next stage, the mice were anesthetized by chloroform, and their organs were removed by careful dissection, exposed to physiological serum for three seconds, and dried on paper for three seconds. Finally, the organs were located separately on glass for weighing.
Data analysis was performed in SPSS version 16, and the plots were illustrated in the Excel software. Data were expressed as mean and standard error of mean (SEM), and the comparison of the test and control groups was performed using t-test at the significance level of P<0.05.

Results

Spleen Weight

The mean weight of the spleen in the control group was 0.25±0.01 grams, while it was 0.11±0.008 grams in the electromagnetic exposure group, and a significant difference was observed between the groups in this regard (P<0.05). In addition, the mean weight of the spleen in the group with electromagnetic exposure and vitamin C supplementation was 0.16±0.01 grams, which indicated a significant difference with the exposure only group (P<0.01).

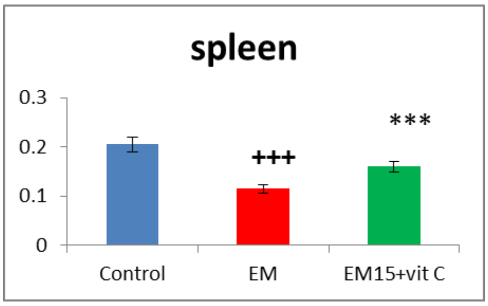

Figure 1. Comparison of Weight of Spleen (g) between Control, Electromagnetic Exposure (Hz), and Electromagnetic Exposure with Vitamin C Supplementation Groups

Kidney Weight

The mean weight of the kidneys of the animals in the control group is 0.21±0.007 grams, while it was estimated at 0.14 ±0.007 grams in the electromagnetic exposure group, denoting a significant difference between the groups in this regard ($P<0.05$). Moreover, the mean weight of the kidneys in the electromagnetic exposure group with vitamin C supplementation was calculated to be 0.19±0.02 grams, indicating a significant difference compared to the exposure only group ($P<0.01$).

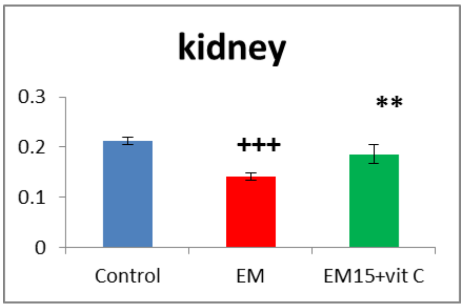

Figure 2. Comparison of Weight of Kidneys (g) between Control, Electromagnetic Exposure (Hz), and Electromagnetic Exposure with Vitamin C Supplementation Groups

Liver Weight

The mean weight of the liver in the animals of the control group was 2.15±0.07 grams, which it was measured at 1.23±0.03 grams in the electromagnetic exposure group, with a significant difference observed in this regard ($P<0.005$). On the other hand, the mean weight of the liver in the animals with electromagnetic exposure and vitamin C supplementation was 1.64±0.14 grams, which indicated a significant difference with the exposure only group ($P<0.01$).

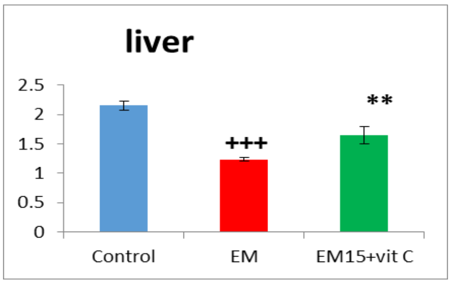

Figure 3. Comparison of Liver Weight (g) between Control, Electromagnetic Exposure (Hz), and Electromagnetic Exposure with Vitamin C Groups (+++Significant difference between control and electromagnetic exposure groups at $P<0.01$; ***Significant difference between electromagnetic exposure and electromagnetic exposure with vitamin C groups at $P<0.01$)

Discussion and Conclusion

Effects of 50 Hz/4 mT Electromagnetic Exposure on the Spleen Weight of Mature BALB/c Male Mice

According to the results of the present study, 50 Hz/4 mT electromagnetic exposure reduced the spleen weight of the mature BALB/c male mice with a significant correlation in this regard. According to Loui Monfared et al., exposure to 915 Hz mobile phone electromagnetic waves for 60 consecutive days decreased lymphocyte population and size of the lymphoid follicle, while increasing neutrophils, monocytes, and IgA levels. The mentioned research also indicated the decreased size, number, and diameter of the viable centers in the lymphoid follicle, follicular area of the lymphocytes, and spleen arterial sheath, as well as the significant reduction of the absolute/relative weight of the spleen in the subjects exposed to mobile phone waves [20]. Researchers have also observed that 20 days of exposure to mobile phone waves in NMRI mice reduced the spleen cells, including T lymphocytes. According to Golestanian and Parivar, the histological study of spleen tissue revealed that the number of megakaryocytes and white pulp diameter increased in 50-100 Gauss fields [21], which is in contrast with our findings. Furthermore, exposure to mobile phone waves has been reported to alter the functionality of spleen cells (e.g., increased cytotoxicity) in rat models [22].

The results obtained by Lino et al. regarding the effects of electromagnetic waves on the embryonic hematopoietic system showed that these waves increased the *erythrocyte sedimentation rate*, which in turn decreased hematocrit [23]. On the other hand, the prevalence of lymphoma and leukemia has been reported to be relatively high among the soldiers who have been permanently exposed to electromagnetic waves [24]. In a study in this regard, Savitz investigated cancerous children and reported that the prevalence of neoplasms was higher in the children with long-term exposure to low-frequency electromagnetic waves, and lymphoid neoplasms were observed to be more prevalent [25]. In addition, Baharara et al. reported the impact of simulated mobile phone waves on the hepatocytes and spleen cells of mice embryos [26].

Effects of 50 Hz/4 mT Electromagnetic Exposure on the Spleen Weight of Mature Male BALB/c Mice Receiving Vitamin C

According to the current research, 50 Hz/4 mT electromagnetic waves increased the spleen weight of the mature male BALB/c mice receiving vitamin C, and the changes in this regard were considered significant. Some studies have also indicated that electromagnetic waves may enhance cell proliferation [27]. The significant increase in the spleen weight could be attributed to the apoptosis inhibitory properties of vitamin C and role of this vitamin in the induction of cell proliferation [28].

Effects of 50 Hz/4 mT Electromagnetic Exposure on the Liver Weight of Mature Male BALB/c Mice

According to the results of the present study, 50 Hz/4 mT electromagnetic waves increased the liver weight of the mature male BALB/c mice, and the changes in this regard were considered significant. In another study, Rajaee and Mohamadian reported that long-term exposure to electromagnetic waves increased the number of the Kupffer cells in the liver [29], which is inconsistent with our findings. On the other hand, Lahijani et al. evaluated the effects of 50 Hz electromagnetic waves on the chicken embryo (white leghorn) before incubation via electron/light microscopy, observing that the electromagnetic waves caused liver cysts with fibrotic bands, sever obstructive hepatitis, and edema. The mentioned study also indicated increased membrane damages and electromagnetic damages [30]. In the current research, hepatic cysts were observed as well.

According to the study by Babayi et al., exposure to a weak electromagnetic field reduced megakaryocytes and liver volume although the reduction was not considered significant; this is inconsistent with the current research. With regard to cell death induction by electromagnetic waves in different cells, previous findings could provide insight into some cues of death cell induction due to the impact of byproducts on the cell cycle. Previously, these products were known as stress proteins, while they have recently been identified in several cells with various functions. However, the reduction of the liver volume in the present study may be associated with cell cycle inhibition with an apoptotic nature. Furthermore, the size reduction of the mature megakaryocytes that were unable to proliferate could be due to the fact that cell death by the waves caused no changes in the cell cycle of a necrotic nature [30].

Effects of 50 Hz/4 mT Electromagnetic Exposure on the Liver Weight of the Mature Male BALB/c Mice Receiving Vitamin C

According to the results of the present study, 50 Hz/4 mT electromagnetic waves increased the liver weight of the mature male BALB/c mice receiving vitamin C, and the changes in this regard were considered significant.

Effects of 50 Hz/4 mT Electromagnetic Exposure on the Kidney Weight of Mature Male BALB/c Mice

Our findings indicated that 50 Hz/4 mT electromagnetic waves decreased the kidney weight of the mature male BALB/c mice, and the changes in this regard were considered significant. According to the literature, exposure to alternate 0.3-0.5 T electromagnetic waves for 4-24 hours has hazardous effects on the kidney cells, including the pyknotic nucleus of the tubular/glomerular cells and cytoplasm shrinkage of the eosinophil; these effects have been reported to be more significant in the cortex than the medulla [31]. In a research, 0.5 mT electromagnetic waves were observed to influence the death/proliferation of kidney cells as shown in the cytometry and morphological analysis. At this intensity, the regular reduction of apoptosis and increase in the necrotic cells were reported as well. The effects of electromagnetic waves depend on the cell type (nephropathogenic cells in the kidneys) [32].

According to the literature, electromagnetic waves could affect intrarenal circulation, which may lead to hemodynamic abnormality, stromal edema, tubular dystrophy, interstitial inflammation, and glomerular sclerosis [33]. The study by Mozaffari and Dezfulian showed that exposure to 0.1 mT electromagnetic waves for one month affected the kidney volume, while also exerting histopathological effect on the kidney tissue. Notably, these effects may vary at different intensities [34].

Effects of 50 Hz/4 mT Electromagnetic Exposure on the Kidney Weight of Mature Male BALB/c Mice Receiving Vitamin C

According to the findings of the current research, 50 Hz/4 mT electromagnetic waves increased the kidney weight of the mature male BALB/c mice receiving vitamin C, and the changes in this regard were considered significant. However, conflicting results have been proposed regarding the effects of electromagnetic fields on the liver and spleen weight. The results of the present study demonstrated that electromagnetic fields could significantly decrease the liver and spleen weight, and receiving appropriate doses of vitamin C may be therapeutic and alleviate the effects of 50 Hz/4 mT electromagnetic waves on mature male BALB/c mice. Therefore, it could be concluded that exposure to electromagnetic waves decreases the kidney weight significantly, which could be attributed to the reduced cell division and apoptosis inhibition. In such cases, vitamin C supplementation is recommended to minimize the kidney damages induced by electromagnetic waves.

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Conflicts of interest

None declared.

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