Research Article

Effect of extremely low frequency electric field on liver, kidney, and lipids of Wistar rats

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Abstract

Background: Increased utilization of electricity and rapid urbanization has caused a greater part of the community to be exposed to extremely low frequency (ELF) fields. Detrimental effects of power frequency fields produced by man-made sources have been notified during the last few decades.

Objective: To detect whether any plausible link exists between exposure to ELF electric field and human health. The effects of ELF electric field (ELF-EF) on liver, kidney, and lipid profiles of Wistar rats were explored.

Materials and Methods: Eighteen Wistar rats with an average age of 2 months were divided into two experimental groups and a control group. The animals were exposed to an electric field of 1.5 kV/m. The first group was exposed for 6 h/day for five consecutive days. The second experimental group was exposed for 6 h/day for 10 consecutive days. Biochemical levels at the end of each exposure period were measured.

Result: The exposure to ELF-EF significantly increased the serum urea and SGOT levels ($p < 0.05$). SGPT, total protein, albumin, and globulin levels were elevated significantly after 30 h exposure, which decreased nonsignificantly for an exposure period of 60 h. Lipid levels did not show any significant difference between the groups. Serum urea, SGPT, SGOT, albumin, and globulin levels increased after the exposure.

Conclusion: Strength and exposure duration of EFs were found to play an important role in inducing internal fields and initiating biological variations. Elevation in levels indicates a probable link between the exposure of ELF-EF and liver and kidney function parameters. These results were further used to calculate the human equivalent fields.

KEY WORDS: Extremely low frequency (ELF), health effects, electromagnetic fields (EMF), lipids, liver, kidney

Introduction

Natural electromagnetic environment of the earth helps the species of animals and birds for migration and navigational guidance. Apart from natural electromagnetic environment, man-made electromagnetic environment is also present. Electromagnetic fields (EMF) in this environment are mostly contributed by power frequency fields around electric power generating stations, distribution lines, high voltage (HV) lines, and electric appliances. Increased speed of urbanization and per capita increase in the utilization of electricity is forcing all kinds of living bodies to be exposed to electromagnetic environment created by man-made sources. To meet the increased demands, electric power generation, distribution, and transmission networks are also experiencing a vast boost. Man-made electromagnetic environment is turning dense owing to power frequency nonionizing radiations around different sources. Biological activeness of power frequency fields came forward after a study in USSR substation in 1972, which associated the fields with potential health hazards.[1] Attempts initiated and conclusions drawn after this study, to assess exposure-oriented effects of extremely low frequency electromagnetic field (ELF EMF), were categorical. Further research in bioelectromagnetics put forward that each organ of biological bodies have different electrical properties such as conductivity and relative permittivity. Biological effects in

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a living organism are initiated by electric fields (EFs) induced inside the body owing to external EMFs. Type of source, distance from source, time spent near to source, strength of fields, the orientation and shape of the body, and, in some cases, its size and tissue composition are also the major parameters when evaluating exposure-oriented effects.[2]

There are certain studies that show negative results of exposure to ELF EMFs and health hazards.[3] However, certain studies indicated that exposure to ELF fields can be a matter of health effects. Researchers have shown probable association of childhood leukemia, brain tumor, breast cancer, respiratory problems, and cardiac attacks on exposure to ELF EMFs.[4–7] Intensity of their effects depends on induced current densities in a living organism.[8] At lower level of induced current densities, effects such as electrophosphenes[9] and effects on pineal gland[10] were observed. Variations in cellular properties have been observed at increased level of densities around 100 mA/m^2.[11]

Early rodent and in vitro studies[12] also demonstrated that field exposure can influence synaptic function, conduction velocity in peripheral nerves, neuronal excitability, and the analgesic effects of opiates. The effects caused by exposure to ELF magnetic field have been dealt extensively. There are recent good attempts to find the effects on internal organs,[13] but very limited researchers have paid attention toward the effects on kidney and liver[14,15] functioning of living beings owing to ELF-EF. Study on biochemical changes in liver and kidney exposed to EMF from mobile phones was also done.[16] Most of the studies applied magnetic field stimulation[17–20] or exposure for a shorter period of time.[14] The exact effects of long-term exposure of EFs on body parameters and organs are still being evaluated. There is a wider scope to decide on the exact level of threshold, which would initiate biological changes in living beings. The direction of the EF is an important parameter that has to be taken into consideration during exposure assessment.[14] In the earlier studies, experimentation was carried out with 0.9 and 1.9 kV/m[15] and 0.3 to 1.8 kV/m[14] in both vertical and horizontal directions, respectively. Vertically applied EF was found to be more effective in terms of increase in superoxide dismutase (SOD) and thiobarbituric acid reactive substances (TBARS) levels of plasma, liver, lung, and kidney tissues.[14,15] By conducting research on ELF-EFs, it is even assumed that this work will help to contribute to further avenues of related topics. The WHO have published Environmental Health Criteria: 238, on ELF fields.[21] The objective of this initiative is to make avail a combined platform for extensive literature related to scattered efforts, attempts, and outcomes of studies related to identify the ill effects of ELF EMF. All studies, with either positive or negative effects, need to be evaluated and judged on their own merits and, then, all together in a weight-of-evidence approach. By far, the majority of studies concern the health effects resulting from exposure to power frequency (50–60 Hz) magnetic fields; a few studies address the effects of exposure to power frequency EFs. To our knowledge, extensive study on the chronic effects of long-time ELF-EF exposure on kidney, liver, and lipid metabolism has not been performed. This study analyzed the levels of certain proteins, enzymes, and bilirubin and abnormalities in lipids by measuring cholesterol and triglycerides and that of kidney by creatinine and urea concentration in serum. A scaling factor for the peak EF strength acting on the outer surface of the body is 4.9:1 for humans versus rats.[2] The results obtained by this study were used to determine the functionally equivalent level of external field, which would initiate biological variations in humans.

Materials and Methods

Animals

Eighteen Wistar rats (9 male and 9 female rats) with an average age of 2 months, weighing between 200 and 250 g were purchased from RC Patel Institute of Pharmacy, Shirpur, Maharashtra, India. Of the 18 Wistar rats, three groups were prepared: two experimental groups and a control group; each group had three male and three female rats. All animal care and procedures were in accordance with Institutional Animal Ethical Committee guidelines.

Exposure Procedure

Exposure procedure was carried out with the help of fabricated pilot treatment unit as shown in Figure 1. It consists of two parallel copper plates, 3,060 cm, one was grounded and other energized by an auto transformer. The EF was created by applying 208 V through autotransformer with 0.1386 m separation between copper plates. In order to protect animals from direct contact with upper energized plate, insulation was provided beneath the upper copper plate at a distance of 1 cm. Wistar rats were exposed with generated EF of 1.5 kV/m. Rats in experimental groups were used for exposure, whereas the rats in the control group were not exposed. The first experimental group was exposed for 6 h/day for five consecutive days whereas the second for 6 h/day for 10 consecutive days.

Figure 1: Experimental setup.

Tests

Blood samples were collected by retro-orbital plexus method. Sample collection was done from group 2 after 30 h of exposure, whereas from group 3 after 60 h of exposure to ELF-EF. Serum creatinine and serum urea were checked by Jaffe's method and urease method, respectively. In liver function test, serum glutamic oxaloacetic transaminase (SGOT) and serum glutamic-pyruvic transaminase (SGPT) levels were measured by kinetic method. Diazo method was used for bilirubin total. Globulin and albumin were measured by bromocresol green (BCG) dye method. Serum cholesterol was measured by peroxidase–antiperoxidase (PAP) method, whereas Trinder method was used to measure triglycerides and HDL cholesterol. All these tests were carried out in coordination with RC Patel Institute of Pharmacy (Shirpur) and Dr. Ulhas Patil Medical College and Hospital (Jalgaon).

Statistical Analysis

Results were evaluated by analysis of variance (ANOVA). A one-way ANOVA was conducted to determine whether parameters are varied as a function of exposed EF. Results were expressed as the mean ± standard error mean (SEM). Differences among the groups were assessed by Dunnett's multiple comparison test using Graph Pad prism, version 5.03. A statistical *p* value less than 0.05 was considered significant for indicating variation, which was owing to real effect rather than by chance.

Result

Kidney Function Test

There were no significant difference between experimental groups and control group for serum creatinine [Figure 2], during the whole period. Moderate increase is observed in serum creatinine after the first phase of exposure period, i.e., at 30 h, whereas it was decreased a bit after the second phase of exposure, i.e., at 60 h. Serum urea showed a significant difference between the control and experimental groups [Figure 2]. The first phase resulted in small increase with no significance, whereas level showed a significant increase at 60-h exposure.

Liver Function Test

No significance was observed for bilirubin total [Figure 3]. Significant difference was observed between experimental and control groups for SGPT [Figure 3]. SGPT levels showed a significant increase at 30 h and a decrease at 60 h. Significant difference was observed among groups for SGOT [Figure 3]. SGOT levels showed a moderate increase at 30 h, which increased to a greater extent at 60 h. There were significant differences in total protein, albumin, and globulin values [Figure 3]. Albumin showed a transient increase at 30 h that decreased at 60 h when compared with controlled group. Total protein and globulin showed transient increase at 30 h that decreased at 60 h, but higher than the control group.

There were no significant differences observed for total cholesterol, triglycerides, and HDL among groups [Figure 4]. Exposure induced almost no variation in the serum cholesterol levels. Initial decrease in triglyceride was observed at 30 h that increased at 60 h. A minor increase in HDL cholesterol level was observed at 30 h that almost reinsisted to initial value at 60 h. Strength and exposure duration of EFs was found to be playing an important role in inducing internal fields and initiating biological variations. The exposure to ELF-EF significantly increased the serum urea and SGOT levels ($p < 0.05$). SGPT, total protein, albumin, and globulin levels were elevated significantly after 30-h exposure, which decreased nonsignificantly for exposure period of 60 h. Lipid levels did not show any significant difference between the groups.

Discussion

Extensive in vivo and in vitro efforts were carried out to determine the biological effects of power frequency EMFs on

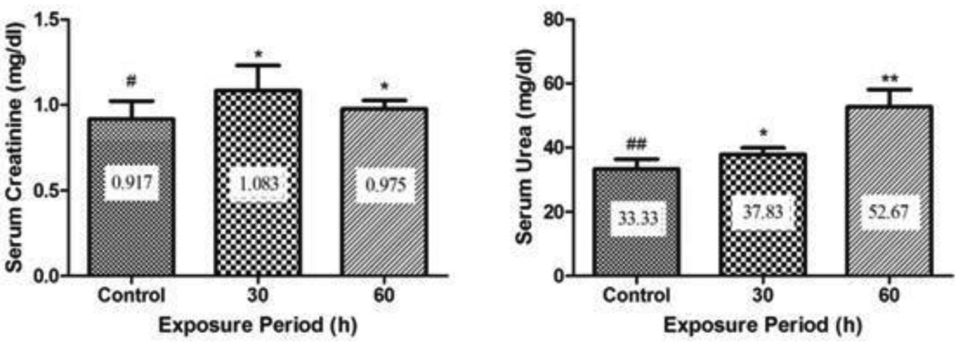

Figure 2: Serum creatinine and serum urea levels in controlled group, 30 h and 60 h exposed groups.

Vertical bars represent SEM, values in text box indicates mean values, while * specify nonsignificance ($p > 0.05$), ** specify significance ($p < 0.05$) to control group, # above control specify nonsignificance ($p > 0.05$) between the groups, and ## above control specify significance ($p < 0.05$) between the groups.

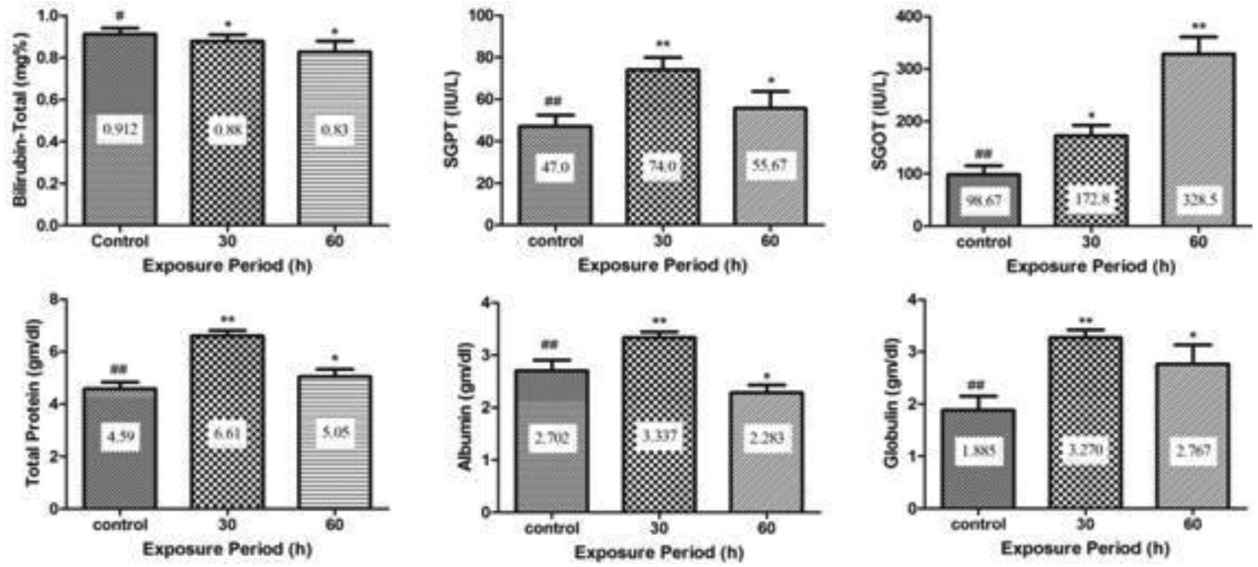

Figure 3: Bilirubin-total, SGPT, SGOT, total protein, albumin, and globulin levels in controlled group, 30-h and 60-h exposed groups.

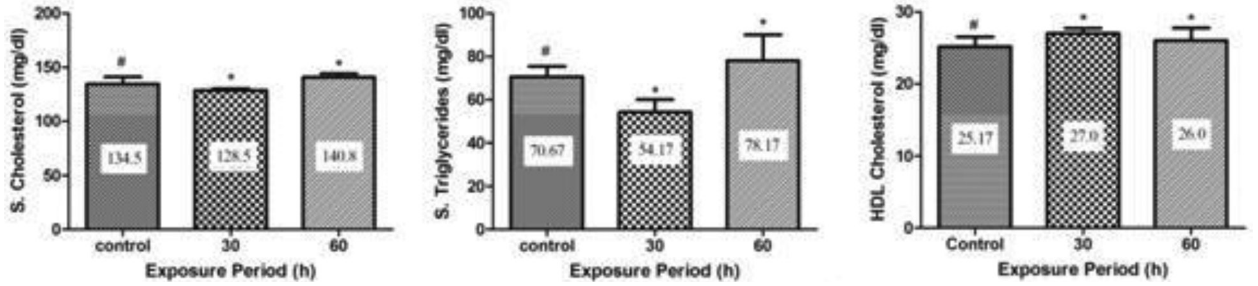

Figure 4: Serum cholesterol, Serum triglycerides, and HDL cholesterol levels in control group and in 30-h and 60-h exposed groups.

living beings. Earlier categorical efforts are now turning out to be organ specific.[22] Earlier studies to evaluate the exposure effects showed that ELF EMF exposure can be an initiator of physiochemical changes, which may further trigger biological changes.[23] Cucullo et al.[24] reported that EFs impact cellular functions by activation of ion channels or by interfering with cell membrane integrity. These changes may affect some biochemical processes and result in changing some serum biochemical parameters and enzymes.[25] This work was carried out to establish whether ELF-EFs generated around power frequency sources have an influence on liver, kidney, and lipid metabolism parameters. Liver and kidney are the two major organs, which process and eliminate any hazardous or toxic substances absorbed in the body. These two organs also regulate the major biological functions associated with healthy life in a living organism. We evaluated liver function by measuring the levels of certain proteins, enzymes, and bilirubin, abnormalities in lipids by measuring cholesterol and triglycerides, and that of kidney by creatinine and urea concentration in serum. Our results showed a significant increase in serum urea ($p < 0.05$) and an increase level of serum creatinine. This is in agreement with Hashem and El-Sharkawy[26] for mice exposed to EMF of 2 mT, 50Hz. On the contrary, a study carried out by Salem et al.[27] on rats exposed to SMF of 128 mT, observed no alterations in the said levels. These results may be owing to the renal dysfunction associated with congestion of renal blood vessels, contracted glomeral tufts of some glomeruli, and focal leukocyte aggregation by pathologic examination.[26] Ibrahim et al.[18] reported an increase in serum bilirubin for rats exposed to 50 Hz magnetic fields; contradictorily, we observed no significant effect on serum bilirubin levels in rats exposed to 50 Hz EF. The levels of albumin, globulin, and total protein after exposure showed significant increase after 30 h when compared with control, which falls a bit after 60-h exposure. The reported increased levels in our study are in agreement with the results obtained through the studies done by Ibrahim et al.[18] and Hashem and El-Sharkawy[26] However, a significant decrease in the levels of total protein, albumin, and globulins were observed in steelworkers exposed to EMF[23] and in rats exposed to EMF.[28] The elevation in the levels of serum albumin and total protein, may result from the damaged cells, which leak into circulation after exposure.[29] The results in this study showed exposure-initiated activities in the levels of both the liver-associated enzymes: alanine aminotranferease (ALT) and aspartate aminotransferase (AST), which were previously known as the SGPT and the SGOT. Significant increase was observed for ALT after 30 h, which further settled to nonsignificant increase than control at 60 h. Nonsignificant increase in AST after 30 h was raised to significant increase after further exposure to 60 h. Exposing to EMFs in humans or animals resulted in increasing glucorticoids (cortisol), stress oxidative compounds, and produced hipoxy. This is an important reason for increasing the amount of transaminases in experimental groups. Hipoxy production could increase the AST and ALT value in serum, up to thousands of units in liter.[25] Both ALT and AST, as reported, are specific liver enzymes that increase in hepatic diseases and toxic damage of liver cells.[30] Significant increase in AST when compared with settled value of ALT owing to EF exposure suggested that the liver was not only organ leaking the AST or that this enzyme was released to a higher degree than ALT from the liver.[31] We found no significant difference in serum cholesterol, serum triglycerides, and HDL cholesterol levels. Similar results were recorded by Torres-Duran et al.[32] for cholesterol and triacylglycerol serum levels with contradictory increase in HDL-C. They investigated that an exposure of 60-Hz, 2.4-mT magnetic field did not cause any change in total cholesterol and triacylglycerol serum levels, but with an increase in HDL-C and free fatty acids at 24 and 48 h, respectively. HDL-C showed lower levels at 96 h, after beginning the exposure. However, Chukwuemeka[33] observed significant exposure-associated differences for total cholesterol and triacylglycerols serum levels, with no significant difference for HDL-C concentration. Harakawa et al.[34] recorded fall in the levels of total plasma cholesterol in the EF-exposed group than that in the control group, which is not according to our findings.

Conclusion

The experimental results show that the ELF EF with foregoing intensity affects the levels of serum urea, SGOT, SGPT, albumin, and globulin. It does not result any significant effect on serum creatinine, cholesterol, triglycerides, and bilirubin levels. In conclusion, our results can be used as a supportive indication of probable role of ELF EF in determining the exposure-oriented effects on liver and kidney functions. The results described here can be tested in other exposure scenarios too. The findings described above could be useful in establishing the link between ELF exposure and health effects. For the exposed unperturbed EF of 1.5 kV/m, the surface EF on rats is 5.5 kV/m, this field is initiating the effect, which is analogous to surface EF of 27 kV/m in humans.

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