MAGNETIC FIELDS HAVE AN EFFECT ON ANTIOXIDANT DEFENSE SYSTEM IN HEART TISSUE

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Abstract: The aim of this study was assessed to evaluate the influences of in vivo exposures to ELF magnetic fields on glutathione (GSH) levels of heart tissue in guinea pigs. Fifty seven, 10 – 12 weeks old, male guinea pigs were used in this experiment. The subjects were divided into one control group and six experimental groups which were exposed to 50 Hz magnetic fields of 10 Gauss (G), 20 G and 30 G with the exposure periods of 4 hours /day and 8 hours/day for 5 days. GSH levels of heart tissue were determined according to the Aykaç et al.'s spectrophotometric method. Mann Whitney-U test was applied for statistical analysis. Mann Whitney-U test was applied for statistical analysis.

GSH levels in heart tissues of guinea pigs exposed to the magnetic fields of 20 G and 30 G were found increased with respect to the controls for both of the exposure periods. For 10 G and 4 hours/day GSH levels increased whereas decreased for the 8 hours/day of exposure periods.

The decrease in GSH level for 10 G magnetic field and 8 hours/day of exposure times and the increase for 4 hours/day were found statistically significant. The increases in GSH levels for magnetic fields of 20 G and 30 G and 4 hours/day of exposure periods and 20 G and 8 hours/day of exposure times were found statistically significant except for 30 G and 8 hours/day of exposure times.

Introduction

Epidemiological studies have shown that there is link between occupational exposure to extremely low frequency magnetic fields and certain cancers such as leukaemia and brain tumors [1-11] It has been known that magnetic fields cause an increase in free radical activity in living organisms [12]. Free radicals are very reactive and unstable molecular species that can initiate chain reactions to form new free radicals [13]. Free radical formation induces changes in enzymes activity, gene expression, alteration of membrane structure and DNA damage [14].

Oxidative stress refers to an imbalance between the intracellular production of free radicals and the cellular defense mechanism. It is due to reactive oxygen species (ROS) generated either by the cellular metabolism. Some ROS are extremely reactive and interact with all the macromolecules including lipids, nucleic acids and proteins. Cells have numerous defense systems to counteract the deleterious effects of ROS. Proteins and small molecules specifically eliminate ROS when they are formed. There are numerous small molecules in the cell such as glutathion, α-tocopherol, vitamines A and C, melatonin, etc. which are antioxydant molecules [12-15].

However, enzymatic and non-enzymatic antioxidants are responsible for scavenging of free radical in body, but at some circumstances, body is insufficient to scavenge them under increased free radical formation.

Antioxidants are increasingly being recognized as important health promoters in conditions such as oxidative stress [16]. Antioxidants are able to reduce the effects of free radicals formed in body. Glutathione is one of the important antioxidant in body defense system for free radical.

Although the muscle cells of the heart consist of an electrical network driven by pacemarker cells situated in the sino atrial nod, the heart demonstrates sensitivity to weak ELF electric fields. It has been known that exposure to ELF might cause some adverse effect on cardiac function has been expressed for many years [17]. However, an acceptable hypothesis has not yet been proposed.

These controversial results have been thought scientist further research on heart tissues the effect of various magnetic fields exposure

Under these findings**,** we examined the effect of various MFs on heart tissue damage of reactive oxygen species (ROS) by measuring GSH levels, which is an important antioxidant.

Materials and Methods

The experimental protocol was reviewed and approved by the Laboratory Animal Care Committee of Gazi University (Report no: 36-7838). In this study, a total of 57 male, 250-300 g weighted (10-12 weeks aged) guinea pigs were used. The animals were fed standard pellet food and kept in the laboratory at a room temperature of 23±0.2 °C, a day and night cycle of 12 hours and ambient geomagnetic field of 0.3 G.

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Magnetic fields were generated using two pairs of circular Helmholtz coils. Coil pairs of Helmholtz configuration were used in the vertical manner. Forty eight guinea pigs were housed in the centre of the Helmholtz coils, 2 per plastic cage and were exposed to 50 Hz, 10 G, 20 G and 30 G fields with the exposure periods of 4 hours/day and 8 hours/day for 5 days. Animals were housed pairly in 26 x 22 x 10 cm^3 plastic cages positioned at the center of the energized Helmholtz coil during experiments, to avoid any distortion of the generated magnetic fields. Nine subjects were handled in an identical manner with the exposed animals in the same laboratory. They were housed at the center of the Helmholtz coils without being exposed to any magnetic fields and used as control. To control possible variation in responses due to circadian rhythm, daily exposure periods of 4 hours and 8 hours were chosen between 8:00-12:00 a.m. and 8:00 a.m. - 4:00 p.m. respectively.

After completion of the experiment, the animals were sacrificed by ether inhalation in a closed box, then lung and kidney tissues were dissected out immediately. They shocked by liquid nitrogen and stored in deepfreeze at -40°C until performing the analysis of contents.

Determination of GSH Levels of Heart Tissue

The GSH levels were determined by modified Aykaç et al. [18] method. Tissues were homogenized in nine volumes of cold 10 % TCA solution and the homogenate was centrifuged for 15 min. at 3000xg at 4°C. The supernatants were transferred to glass test tubes. 0.5 ml of supernatant was added to the 2 ml of 0.3 M Na_2HPO_4 $2H_2O$ solution. A 0.2 ml solution of dithiobisnitrobenzoate (0.4 mg/ml 1 % sodium citrate) was added, and absorbance at 412 nm was measured immediately after mixing. The GSH levels were calculated using an extinction coefficient of 13.600 mol^{-1} cm^{-1}.

Statistics

Statistical analyses were carried out using SPSS software (SPSS Inc., Chicago, USA). The P value was considered significant at $P<0.05$.

Mann Whitney-U and Kruskal-Wallis tests were used in statistical analysis. Comparisons between exposed groups and controls were made by using Mann Whitney-U test while magnetic fields of 10 G, 20 G and 30 G were compared with Kruskal-Wallis test with respect to exposure periods of 4 hours and 8 hours.

Results

GSH levels were investigated in heart tissues of guinea pigs in the presence of 50 Hz, 10 G, 20 G and 30 G magnetic fields with the exposure periods of 4 hours/day and 8 hours/day for 5 days.

Increases in GSH levels for the exposure period of 4 hours/ 5 days were found statistically significant in heart tissues under the effects of the 50 Hz magnetic fields of 10 G, 20 G and 30 G with respect to controls..

By the magnetic fields of 10 G, 20 G and 30 G applied for 4 hours, heart tissues GSH levels were measured 3.86±0.29 µmol/g tissue, 3.43±0.37 µmol/g tissue and 3.49±0.27 µmol/g tissue respectively. GSH levels in control group was found as 2.01± 0.15 µmol/g tissue.

For 10 G, 20 G and 30 G applied for 8 hours, heart tissues GSH levels were measured 1.47±0.13 µmol/g tissue, 3.14±0.15 µmol/g tissue and 3.44±0.25 µmol/g tissue respectively. GSH levels statistically increased only for 50 Hz, 20 G fields with the exposure periods of 8 hours/day in heart tissue.

All of the GSH values given in Figure 1 are mean±standart error of mean.

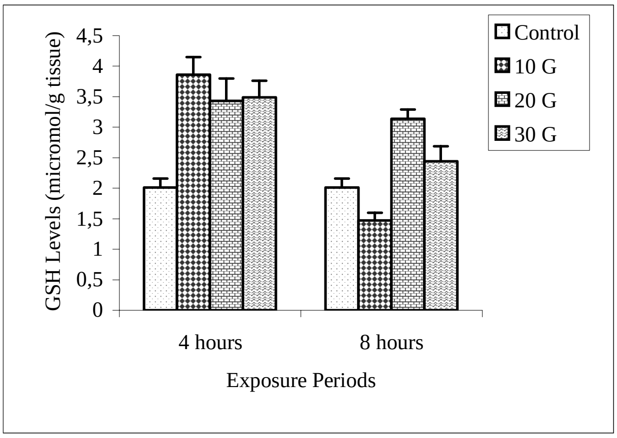

Figure 1. GSH levels of heart tissues of guinea pigs exposed to magnetic fields and controls.

Discussion

All the members of modern societies constantly live in MF and waves whose intensities are much higher than those found in the nature. These negative effects of MF on human health would be continuing as a controversial.

In this study, to obtain data related with the effects of magnetic fields on free radical metabolism, we measured hearth tissue GSH levels on guinea pigs which were exposed to 50 Hz magnetic fields of 10 G, 20 G and 30 G with the exposure periods of 4 hours /day and 8 hours/day for 5 days.

It is hypothesized that modification of structural and functional properties of the membrane due to inhibition of free radical processes increases functional activity of Ca-pump in sarcoplasmic reticulum [19].

It has been known that the heart and pineal gland were not sensitive to the direct effects of weak ELF fields [20]. There is not much clear evidence that 50-Hz EMF is associated with other neurodegenerative or cardiovascular diseases [21]. Our results indicated that GSH levels increased in the heart tissue of guinea pigs which exposed to 50 Hz magnetic fields of 10 G, 20 G and 30 G with the exposure periods of 4 hours /day and 8 hours/day for 5 days.

Conclusion

The biological effects of MF remain a matter of debate because of supporting or disproving results on body cells. These discrepancies might at least in part be attributed to experimental factors like intensity, duration and cell type. These results indicate that chronic exposure to various ELF-MF increase GSH levels, which are free radical scavengers in heart tissue.

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