12 Hz electromagnetic field changes stress-related hormones of rat

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ABSTRACT

Due to the increasing presence of electromagnetic radiation in advanced societies, investigation of the effects of this radiation on humans has attracted the interest of many researchers. The non- ionizing radiation can induce numerous effects through biomolecules in the tissue and cells. In the current study, effects of 12 Hz electromagnetic waves investigated on neuronal hormone levels in rat animal model in 1, 3, 7, 14 and 21 days after exposure. The levels of Adrenocorticotropic hormone (ACTH), Adrenaline, melanocortin-2 receptor (MC2R), corticostrone, α1-receptor and D-Glucose were evaluated by ELISA methods. Exposure with 12 Hz electromagnetic waves did not alter in glucose and MC2R levels in rats compared with control groups. Plasma ACTH and adrenaline concentrations as two important stress-related hormones increased significantly 1 and 3 days after irradiation respectively. Also corticostrone and alpha a1-receptor decreased significantly at some days during the period of study in irradiated rats. Our data demonstrated that faced with 12 Hz electromagnetic wave could be considered as a stress inducer and also time of exposure have critical role in stress induce.

Keywords: 12 Hz electromagnetic; waves; adrenaline; d-glucose; corticostrone; MC2R; ACTH; alpha a1-receptor

INTRODUCTION

Extremely low frequency (ELF) waves as one of the most important and effective range of electromagnetic waves in the field of biology and neurophysiology, especially in the last decade have been more attracted of researchers attention [1, 2]. Today, there are wide range of ELF waves in environment [3] and in the other hand, possible applications of radiation in the treating diseases and disorders of the cognitive and behavioral problems [4-6] caused to intensify the need of experimental study about the effects of these waves. ELF electromagnetic fields (ELF-EMF) are non-ionizing and the beam energy is very weak to separate the orbital electrons of atoms [7]. Several previous studies on the effects of non-ionizing electromagnetic fields were carried out and their results showed that the primary objective of this wave is targeting of chemical processes involved in metabolic pathways [8-11]. Effects of electromagnetic fields on the living organisms are a complex phenomenon [12]. The physicochemical activity of electromagnetic fields is including electron, ion, dipole structure of macromolecules and electrical polarization [13]. Other factors may also play a role, such as excitation of molecular, biochemical activation, production of radicals, weakening the chemical bonds, water-blind changes, atomic vibration and modified spin relaxation times of dipoles [14, 15]. These chemical changes can affect the serum biochemical parameters. Many studies have shown that changes in protein levels [16, 17].

The biological effects of electromagnetic fields are presented as a series of interconnected steps. The first step is the exposure to these fields. In the second phase magnetic fields shall react with molecules of biological structures until could change them shape, size, charge or energy and ultimately cause to produces a signal in the cells. If this signal in frequency and time was differentiated with the thermal threshold produced by the continuous movement of charged molecules in the cell, causes to changes in cells behavior which results in a biological effect is visible [18], This effect can be uncomplicated or complicated.

If changes were in the range of normal variation or a combination of physiological compensatory mechanisms, it was without complication, but the changes weren't in the range of natural variation, having a problem.

Body cells, to reduce the side effects of this field use protections such as increased levels of epinephrine and melatonin [10]. Although the exact mechanism of action of these compounds in reducing the side effects are still not well known. But it is believed that both factors reduce the adverse effects of free radicals [19].

In the present study, the effect of ELF-EMF exposure on some of the neuronal functions by evaluation of neuronal hormones such as Adrenocorticotropic hormone (ACTH), Adrenaline, melanocortin-2 receptor (MC2R), corticostrone, α1-receptor and D-Glucose by Elisa in 1, 3, 7, 14 and 21 days during the period of study.

MATERIALS AND METHODS

Animals

Twenty male Sprague-Dawley rats with weighing 250-300 g were purchased for this study (n=10/group, Razi Institute, Tehran, Iran). The rats were housed in a animal room during 2 weeks for adaptation under a 12 h light: 12 h dark cycle and controlled temperature (21 to 25˚C) with free access to food and water. In the present study, All procedures for this investigation were performed in accordance with the Ethics Committee of Baqiyatallah (a.s.) University (357: November 2000, Tehran, Iran) guidelines. All experiments were carried out between 12:00 and 14:00 h and each rat was examined only once.

Shielded room

As it is reported in previously [20], to prevent the possible interference of foreign interference wave with testing 12Hz, a room space is designed and all parts of the room , including the ceiling and walls, windows , even the smallest openings completely covered and sealed with aluminum foil (0.4 mm diameter). For further confirmation of this step the room was checked by a wave detector and results were confirmed the lack of effective radiation in the shielded room. The electromagnetic generator antenna symmetrically mounted in the box above (for uniform wave radiation). All the conditions for the control group were considered, except irradiation.

Experimental design

In the present study, 12Hz frequency of EMF was irradiated to treated groups once a day with 75mW and 0.1 mT during 21 days. Animals were sacrifice to prepare blood sample to evaluation of neuronal hormones and hormone receptors on days 1, 3, 7, 14 and 21 using ELISA kit.

Hormonal analysis

ACTH, Adrenaline, MC2R, corticosterone, α1-receptor and D-Glucose were measured using the appropriate kites (all from CUSABIO CO., Japan) and a micro-plate reader (ELx-800, BIO-TEK instruments, Winooski, VT, USA). Blood samples were collected in tubes content 5% EDTA and then were centrifuged in 4 °C for 5 min in 3000 RPM. The supernatant was collected for evaluation by ELISA assay.

Statistical analysis

For statistical analysis, Two-Way ANOVA was used by time and frequency as the factors. Further analyses for individual between-groups comparisons were carried out with post hoc Tukey's test. Data were showed as the mean ± SD. In all comparisons, $P<0.05$ was considered to be statistically significant. All statistical analyses were conducted with SPSS software, version 11.0 (SPSS, Chicago, IL, USA).

RESULTS

ACTH

As showed in figure 1, significantly increase of ACTH level were seed only one day after irradiation with 12 Hz ELF-EMF in treated group compared to control and their level in other days was not changed.

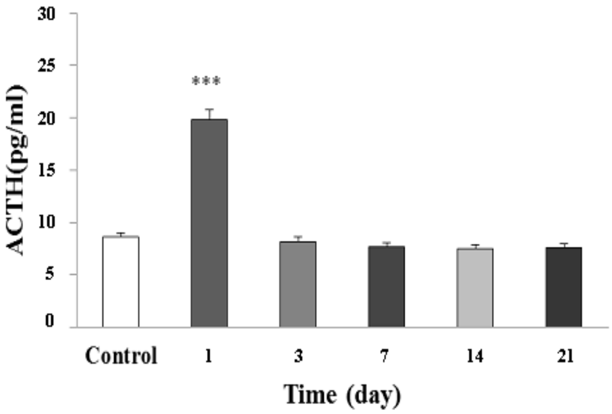

Figure 1. Changes in ACTH levels in rats exposed for 21 days to 12 Hz electromagnetic waves. Asterisk indicates a significant difference between P < 0.05, two asterisks P < 0.01 and three asterisks P < 0.001 between the two groups.

Adrenaline

First day after irradiation increase of adrenaline level observed in the rats and only day 3 this increase was significant in comparison to control group (Figure 2). Also, after day 3 decreasing trend of adrenaline detected in irradiated rats.

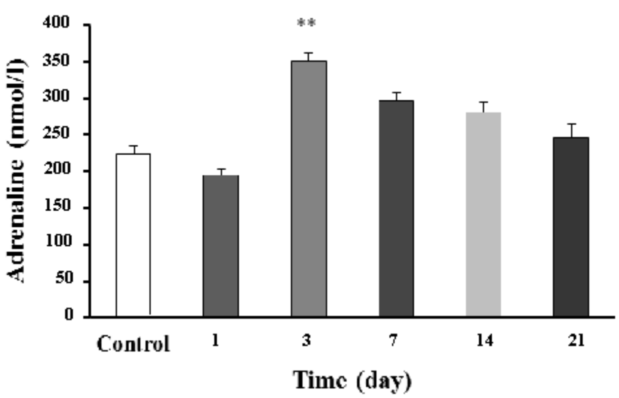

Figure 2. Changes in adrenaline levels in rats exposed for 21 days to 12 Hz electromagnetic waves. Asterisk indicates a significant difference between P < 0.05, two asterisks P < 0.01 and three asterisks P < 0.001 between the two groups.

MC2R

Alterations of MC2R in rats after irradiation were not significant in comparison to un-irradiated rats (Figure 3) and could be included that 12 Hz electromagnetic waves weren't affects on this hormone receptor.

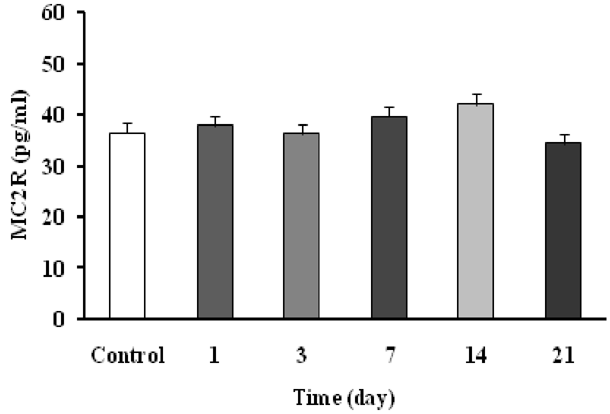

Figure 3. Changes in MC2R levels in rats exposed for 21 days to 12 Hz electromagnetic waves. Asterisk indicates a significant difference between P < 0.05, two asterisks P < 0.01 and three asterisks P < 0.001 between the two groups.

Corticosterone

Evaluation of corticostrone hormone in rats was exposure by 12 Hz electromagnetic waves showed that this hormone level significantly decreased only day 1 after irradiation (Figure 4). In days 3, 7 and 14, this decrease wasn't significant and in day 21 level of this hormone increased in comparison to control group but wasn't significant.

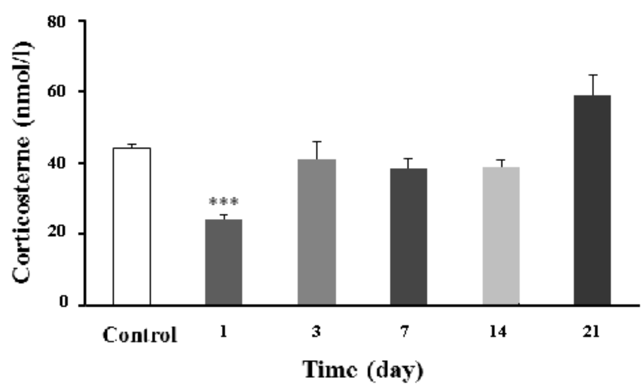

Figure 4. Changes in corticostrone levels in rats exposed for 21 days to 12 Hz electromagnetic waves. Asterisk indicates a significant difference between P < 0.05, two asterisks P < 0.01 and three asterisks P < 0.001 between the two groups.

Alpha A1-receptor

As showed in figure 5, significantly decrease in alpha a1-receptor level was observed on day 1 in 12 Hz electromagnetic wave irradiated rats compared with control group. In days 3 and 7 not any change displayed while in days 14 and 21 level of this receptor increased but this change wasn't significant.

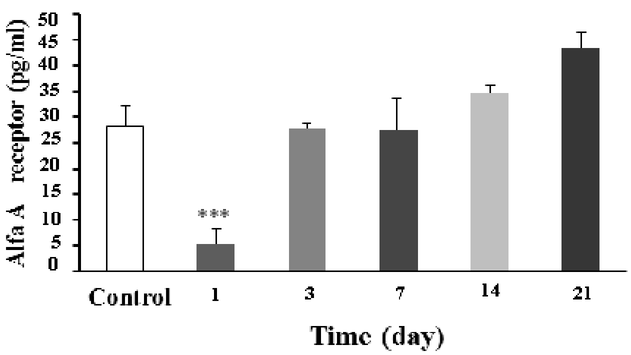

Figure 5. Changes in alpha a1-receptor levels in rats exposed for 21 days to 12 Hz electromagnetic waves. Asterisk indicates a significant difference between $P < 0.05$, two asterisks $P < 0.01$ and three asterisks $P < 0.001$ between the two groups.

Glucose

As displayed in figure 6, glucose level not changed significantly in irradiated rats compared with control groups. Although, this alteration in day 3 was more than other days in irradiated rats but this change wasn't significant in comparison to control group.

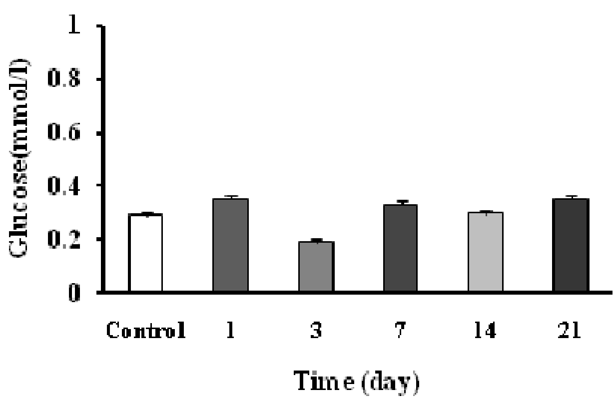

Figure 6. Changes in D-Glucose levels in rats exposed for 21 days to 12 Hz electromagnetic waves. Asterisk indicates a significant difference between $P < 0.05$, two asterisks $P < 0.01$ and three asterisks $P < 0.001$ between the two groups.

DISCUSSION

Many quantitative studies were done about ELF-EMF effects on the pituitary hormones and other endocrine glands hormones [21-23]. The major hormones that were reviewed in ELF-EMF studies, including several hormones that were involved in the physiology and growth, especially thyroid-stimulating hormone (TSH) and thyroid gland that controls the release of thyroxine, Adrenocorticotropic hormone (ACTH) controls the function of the adrenal cortex, especially cortisol release (similar corticosterone in rats), and growth hormone (GH), which affects the growth of the body [24]. Other hormones released by the pituitary were also studied, especially follicle stimulating hormone (FSH) [25], luteinizing hormone (LH) [26] and prolactin [27] that are important for sexual and reproductive functions. Both FSH and LH hormones affect on testicular function and testosterone release. In another study was done on workers exposed to magnetic fields and electric ELF, the levels of ACTH increased when working with leisure time report, the authors emphasizes that the results are more dependent on the factors are attributable to work-related than ELF-EMF [28]. The possibility that ELF-EMF could act as stress is was investigated in several studies [29, 30]. In these studies were examined the effects of electromagnetic field exposure on secrete hormones involved in stress responses, especially cortisol, ACTH and corticosterone release from the adrenal cortex. Reports suggest that serum corticosterone levels in young rats immediately increase after the first exposure. But also in some other reports have been suggested that exposure to ELF did not show any effect on ACTH, cortisol or corticosterone levels. In a study reported by Zare et al. showed that electromagnetic radiation causes to decrease in cortisol and glucose levels [31]. Loss can be resulting of hormone synthesized in the kidney, which is controlled by peptide hormones.

Access tissue proteins, releasing amino acids and their metabolism targeting by catabolic activity of glucocorticoids in the liver, but the changes observed in glucose levels can only be related to glycolysis and Glycogenolysis by blocking the conversion of pyruvate to acetyl-CoA. So they admitted that whole-body exposed to a field of 50 Tesla reduce 0.207 Hz may decrease glucose and cortisol levels. There are conflicting reports about the impact of MF or EMF waves on the rate of increase or decrease in blood glucose. The study of Seyednur et al. noted that their findings confirmed an increase in blood glucose levels [32]. In another study Zare et al. were investigated whole-body exposure effects on changes in the level of cortisol and glucose. In this study, the frequencies of 5 and 50 Hz were used within 5 days of radiation doses 2 and 4 hours per day. Their results showed a significant decrease in and glucose levels in the radiation frequency of 50 Hz, 2 hours per day [33].

ELF-EMF potential as a stressor was studied in several studies, including Hackman and Graves in 1981 [34, 35]. In this study the effects of electromagnetic field exposure were studied on the secretion of hormones involved in stress responses, especially ACTH and cortisol or corticosterone release from the adrenal cortex. Results demonstrated that serum corticosterone level was increased immediately after the first exposure in young rats and also reported that exposure to ELF did not show any effect on ACTH, cortisol or corticosterone levels. In Conclusions, Our data demonstrated that after irradiated with 12 Hz electromagnetic waves did not significantly change observed in glucose and MC2R levels in comparison to control groups.

Plasma ACTH and adrenaline levels as two more important stress-related hormones increased significantly 1 and 3 days after irradiation respectively, these results showed that time of exposure have critical role in induce stress.

Also corticostrone and alpha a1-receptor decreased significantly at some days during the period of study in irradiated rats.

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