



Antioxidant Effects of *Allium cepa* and Cinnamon on Sex Hormones and Serum Antioxidant Capacity in Female Rats Exposed to Power Frequency Electric and Magnetic Fields

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Abstract

Objectives: In this study we investigated whether 50 Hz power frequency electric field of 1.01×10^3 V/m and the magnetic field flux density of 3 mT have effect on serum sex hormones and antioxidant capacity and if *Allium cepa* and cinnamon can moderate potential adverse effects of power frequency electromagnetic exposure.

Materials and Methods: Forty female Wistar rats, weighting 220 ± 10 g were used for this study. Rats were allocated randomly to four groups. The first group (the control group) received 3 cc normal saline (0.9%) daily and was treated for 6 weeks. The second group was exposed to power frequency of 3 mT for 4 h/day, 7 days/week for 6 weeks. The third group received *Allium cepa* (3 cc onion juice) and cinnamon (75 mg/kg body weight) daily for 6 weeks by Gavage method. The fourth group was exposed to power frequency EMF of 3 mT for 4 h/day, 7 days/week and received *Allium cepa* (3 cc onion juice) and cinnamon (75 mg/kg body weight) daily for 6 weeks by Gavage method.

Results: Levels of luteinizing hormone (LH), follicle-stimulating hormone (FSH) and estrogen, significantly decreased in second group that exposed to power frequency EMF of 3 mT and 10^3 V/m ($P < 0.05$) and modified in exposed rats which received *Allium cepa* and cinnamon ($P < 0.05$). Also, TAC levels were significantly decreased in exposed rats and were moderate in exposed rats received *Allium cepa* and cinnamon ($P < 0.05$).

Conclusion: Power frequency electromagnetic field could adversely affect sex hormones and total antioxidant capacity (TAC) levels in exposed rats and *Allium cepa* and cinnamon could prescribed as an effective pharmacological supplement to moderate exposure degenerative effects.

Keywords: Extremely low frequency, Electromagnetic fields, Follicle stimulating hormone (FSH), Luteinizing hormone (LH), Estrogens

Introduction

In addition to the background radiation of natural electromagnetic fields of the earth, human being is at the risk of manmade sources of electromagnetic field. Recently, with rapid development of new technologies, public concerns on potential effects of power frequency electromagnetic field on reproduction and fertility have been increased. Low electromagnetic fields of 1-300 Hz are commonly use and occur in power transmission lines, rail way power supply, transformers and in household power supply and devices (1,2).

According to International Commission on Non-Ionizing Radiation Protection (ICNIRP) guideline, direct interact of external alternating low frequency magnetic field (power-frequency) could induce electric fields and circulating electric current flow within the human body. Also, the direct interaction of external alternative (power-frequency) electric field with the body could induce time-varying surface charge on the body results in induced currents in

body. Each body tissue has finite conductivity. Magnitude of induced electric fields, currents and charges within body and depth of penetration depends on magnitude of the magnetic flux density and frequency of applied extremely low electromagnetic field (3,4). The energy absorption and temperature rise induced by ELF-EMF in body are Inconsiderable. High internal current density results adverse biological effects on body. The induced current density by ELF electric field of 60 Hz 10^3 V/m in waist is 0.427×10^{-3} (5).

In the range of extremely low frequency, electric and magnetic fields are measured separately because, in this range pure electric field E and almost pure magnetic H field could appear in some regions and E and H act independent and we could not reach to peak of both electric and magnetic fields in same location (6).

The International Agency for Research on Cancer (IARC) has classified ELF EMF as a 2B possible carcinogen in humans and less than sufficient in animals in June 2001

Received 3 February 2016, Accepted 1 July 2016, Available online 1 July 2016

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(7). Many studies have shown that exposure to extremely low electromagnetic fields can have adverse effects on reproductive system and tracts, and thus, a relationship between exposure to electromagnetic fields and infertility is conjectured (8).

Evidence showed that balance between reactive oxygen species (ROS) and biological antioxidants plays an important role in women's fertility and reproductive health (9). Electromagnetic fields have adverse effects on biological systems. Oxidative stress occurs as a result of increase in ROS concentrations in cells leading to various pathological damages (10,11).

Cellular metabolisms and other exogenous environmental sources such as electromagnetic fields could cause ROS production in cells. High concentrations of ROS have been implicated to many infertility problems (12).

Dietary and pharmacological supplements were assumed to be helpful to reduce degenerative effects of ROS and free radicals on biological system. Antioxidants are chemical or biological agents that reduce oxidative damages. Antioxidant capacity white *Allium cepa* is rich in phenolic compounds in different layers and has high antioxidant activity (13,14). Anti-oxidative and anti-mutagenic properties of cinnamon as a rich source of polyphenolic compounds have been indicated. Cinnamon could modify oxidative stress induced by different factors and has long been prescribed to cure fertility problems and other diseases (15).

The aim of this study was to evaluate adverse effects of 50 Hz, 3 mT power frequency EMF on serum LH, FSH and estrogen and total antioxidant capacity (TAC) levels and if *Allium cepa* and cinnamon can reduce adverse effects of electromagnetic exposure.

Materials and Methods

Animals

Forty female Wistar rats, weighting 220 ± 10 g were used for this study. Rats were 8-9 weeks old and were quarantined for 1 week maintained under standard conditions with respect to humidity (35% to 60%), illumination (12 hours light and 12 hours by dark fluorescent light controlling) and temperature ($20 \pm 2^\circ\text{C}$).

Rats were allocated randomly to four groups. A control group ($n = 10$) and three treatment groups (total = 30) and $n = 10$ in each.

The first group (the control group) received 3 cc normal saline (0.9%) daily and was treated for 6 weeks.

The second group was exposed to ELF-EMF of 3 mT for 4 h/day, 7 days/week for 6 weeks.

The third group received *Allium cepa* (3 cc onion juice) and cinnamon (75 mg/kg body weight) daily for 6 weeks

by Gavage method.

The fourth group was exposed to ELF-EMF of 3 mT for 4 h/day, 7 days/week and received *Allium cepa* (3 cc onion juice) and cinnamon (75 mg/kg body weight) daily for 6 weeks by Gavage method.

On the 42nd day Blood samples were collected from the tail vein. At the end of the work, animals were sacrificed by cervical dislocation of the neck.

ELF-EMF Exposure Setup

The ELF-EMF exposure device used in this study was consisted of two main parts, the radiation area and a transformer. The radiation area was a cylindrical chamber that an electric and a magnetic field were produced there based on the Helmholtz coils. A pair of cooper coils was placed in radiation area one above the other. Coils were separated by the distant of 50 cm which adjusted to produce a time varying current of 50 Hz results the average ELF electric field of 1.01×10^3 V/m and the magnetic field flux density of 3 mT. Magnetic field strength was 0.24 A/m.

Cages were placed between the coils where a highly homogeneous electric and magnetic field were produced.

The transformer was used to control input and output voltage and current. The stability of voltage and frequency in ELF-EMF exposure system used in this study is similar to voltage and frequency characteristics of power line Distributions. The output voltage was 25 VAC with stability of 10% max.

Electric and magnetic field measured in certain radiuses at 8 locations and the average was reported. To avoid temperature changes, a fan was placed in chamber (radiation area). Fluorescent light distant from the device was 3 m (Table 1, Figure 1).

FSH, LH and Estrogen Hormones Levels Measurement

Serum levels of follicle stimulating hormone (FSH), luteinizing hormone (LH) and progesterone were assayed by two-site chemiluminescence (sandwich) immunoassay (16).

Serum Total Antioxidant Capacity Measurement

Serum TAC measurement is performed by means of a commercial kit (Randox Co., England). In this measurement, incubation of 2,2'-Azinobis-(3-ethylbenzothiazoline-6-sulphonate) (ABTS) with a peroxidase (methmyoglobin) and hydrogen peroxide was done to produce the radical cation ABTS⁺. ABTS⁺ is colorimetrically blue-green, radical cation which is measured at 600 nm (17).

Data analysis

Statistical analysis was done using the Analysis of variance

Table 1: ELF-EMF Exposing Parameters^a

Duration of exposure	Frequency (Hz)	Electric field strength, (V/m)	Magnetic field strength, B (A/m)	Magnetic field strength, H(mT) ^b
4 h/day For 6 weeks	50	$1.01 \times 10^3 \pm 0.05$	24×10^2	3 ± 0.01

^aData is presented as mean \pm SD.

^b $B = \mu_0 H$, $\mu_0 = 4\pi \times 10^{-7} \text{NA}^{-2}$ [magnetic permeability newtons/ampere²].

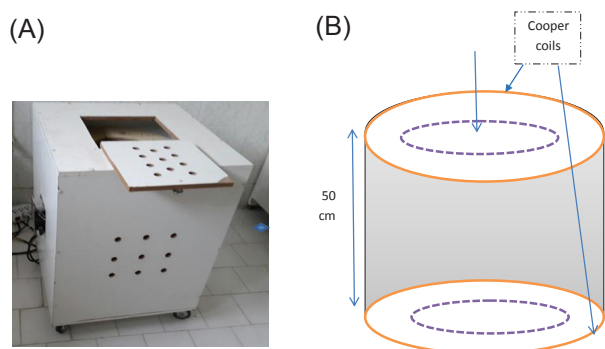


Figure 1. (A) ELF-EMF Producing Device. (B) Diagrammatic Scheme of Radiation Area.

(ANOVA) test for comparison of data in the control group with the experimental groups. The results were expressed as mean \pm SEM (standard error of means). P value less than 0.05 were considered significant.

Results and Discussion

Electric devices which work by electrical power currents are used both in AC and DC mods. According to the guide issued by the ICNIRP, the direct interaction of external time-varying low frequency electric and magnetic fields could cause alternating electric fields and electrical currents inside the body.

At 50-60 Hz, common time-varying (AC) external electric field of 10^3 V/m will generate an average time-varying electric field of 4 mV/m within the body. ELF magnetic field strength (H) inside the body is equal to that of outside body due to equality of magnetic permeability μ of most biological materials with that of free space ($4\pi 10^7$ H/m).

Many studies have been indicated the relationship between ELF EMF and free radicals and ROS production in body (18). ELF EMF exposure may be harmful by impairing the antioxidant defenses (19). Results indicated that extremely low magnetic field of 60 Hz could adversely affect body antioxidant defensive system by ROS production, other than superoxide radicals (20).

Results of TAC Levels Measurement

TAC level in control and EMF groups was 0.77 ± 0.05 and 0.44 ± 0.05 mmol/mL, respectively. Also, in cinnamon & *Allium cepa* and cinnamon & *Allium cepa* + EMF groups, it was 2 ± 0.02 0.61 ± 0.05 mmol/mL, respectively. One-way ANOVA shows significant differences between experimental groups in comparison to control group ($P < 0.05$;

Table 2).

Results of FSH Levels Measurement

FSH level in control and EMF groups was 1 ± 0.22 and 0.6 ± 0.05 nmol/mL, respectively. Also, in cinnamon & *Allium cepa* and cinnamon & *Allium cepa* + EMF groups, it was 2.5 ± 0.05 and 1 ± 0.05 nmol/mL, respectively. ANOVA shows significant differences between experimental groups in comparison to control group ($P < 0.05$; Table 2).

Results of LH levels measurement

LH level in control and EMF groups was 0.6 ± 0.05 and 0.49 ± 0.05 nmol/mL, respectively. Also, in cinnamon & *Allium cepa* and cinnamon & *Allium cepa* + EMF groups, it was 5.5 ± 0.05 and 3.3 ± 0.05 nmol/mL, respectively. One-way ANOVA shows significant differences between experimental groups in comparison to control group ($P < 0.05$; Table 2).

Results of Estrogen Levels Measurement

Estrogen level in control group was 65 ± 0.01 ng/l and in EMF group was 51 ± 0.05 . In cinnamon & *Allium cepa* received group was 83 ± 0.05 and in cinnamon & *Allium cepa* + EMF group was 60 ± 0.05 ng/l. One-way ANOVA shows significant differences between experimental groups in comparison to control group ($P < 0.05$; Table 2). Levels of LH, FSH and estrogen, significantly decreased in second groups that exposed to power frequency EMF of 3 mT and 10^3 V/m ($P < 0.05$) and modified in exposed rats which received *Allium cepa* and cinnamon ($P < 0.05$). Also, TAC levels were significantly decreased in exposed rats and were moderate in exposed rats received *Allium cepa* and cinnamon ($P < 0.05$; Table 2).

Estrogen regulates gonadotropins release. Also, growth of ovarian follicles ovulation and the number of pre-ovulatory follicles are regulated by secretion of LH and FSH. Also, production of estrogen and testosterone depend on high LH secretion. It has been indicated that estrogen has significant antioxidant effects at high concentrations (21-24). Levels of LH and FSH were reduced significantly in rats after six weeks of exposure to ELF-EMF of 50 Hz and 25 μ T. Results show that effects of ELF-EMF exposure on sex hormones are reversible (25). Also other study showed that exposure to 50 Hz ELF magnetic (strength of 5 mT) could affect FSH and LH serum levels (26).

Power frequency EMF exposure may be harmful by impairing the antioxidant defenses. Accurate assessment of

Table 2. Serum Analysis^a

Samples Group	Control	ELF-EMF	Cinamon & <i>Allium cepa</i>	ELF-EMF Cinamon & <i>Allium cepa</i>
TAC mmol/mL	0.77 ± 0.05	$0.44 \pm 0.05^*$	$2 \pm 0.02^*$	0.61 ± 0.05
LH nmol/mL	0.6 ± 0.05	0.49 ± 0.05	$5.5 \pm 0.05^*$	$3.3 \pm 0.05^*$
FSH nmol/mL	1 ± 0.22	$0.6 \pm 0.05^*$	$2.5 \pm 0.05^*$	1 ± 0.05
Estrogen ng/l	65 ± 0.01	$51 \pm 0.05^*$	$83 \pm 0.05^*$	60 ± 0.05

Abbreviations: TAC, total antioxidative capacity; LH, luteinizing hormone; FSH, follicle-stimulating hormone.

*Significantly different at $P < 0.05$ level (compared with the control group).

^aData are presented as mean \pm Standard error (SE).

induced oxidative stress in biological systems play an important role in investigations which conducted to study role of free radical damages in human pathogenesis TAC data obtained from plasma level is conjured as biomarkers of oxidative stress (27).

ROS production could be affect by ELF-EMF exposure. Based on conducted studies oxidative stress could induce female pathogenesis leading to female fertility and sub-fertility. ROS could affect physiological functions of female reproduction system tracts and has been implicated in ovarian follicles apoptosis under influence of external chemical and physical factors (20,28,29).

Based on results, *Allium cepa* and cinnamon could moderate adverse effects of exposure to electromagnetic field on LH and FSH and estrogen level. Hormones levels were significantly increased in exposed rats which received *Allium cepa* and cinnamon.

Conclusion

Power frequency electromagnetic field could adversely affect sex hormones and total antioxidant capacity levels in exposed rats and *Allium cepa* and cinnamon could manuscript as an effective pharmacological supplement to moderate exposure degenerative effects.

Ethical Issues

The ethical committee of Tabriz University of Medical Sciences approved the study.

Conflict of Interests

All the authors declare that there is no conflict of interest.

Financial Support

This paper was based on MSc thesis of Elham mansouri with which was supported by the research grant of Women's Reproductive Health Research Center, Tabriz University of Medical Sciences, Tabriz, Iran, with research code 93/2-4/9 and research ethical code 9315 that was approved in 2014.

Acknowledgments

With special thanks to Women's Reproductive Research Center, Tabriz University of Medical Sciences, Tabriz, Iran for their comments and financial support.

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