

Full Length Research Paper

Antidepressant-like activities of *Ocimum basilicum* (sweet Basil) in the forced swimming test of rats exposed to electromagnetic field (EMF)

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Medicinal use of basil, *Ocimum basilicum*, dates back to ancient times in Iran, China and India. In this study, we aimed to experiment on the antioxidative property of a herbal source (sweet basil) to protect central nervous system against oxidative damages of electromagnetic field (EMF) and its affective sequences. We used forced swimming test to evaluate antidepressant effect of basil extract in 30 albino male Wistar rats that had been exposed to 50 Hz, EMF for 8 weeks. At the end of 8th week, rats which have been feed with basil extract (1.5 g/kg body weight), showed decrease in immobility score ($P < 0.001$) and increase in swimming ($P < 0.001$), but not in climbing scores significantly in comparison with control group. Basil extract can act as serotonergic antidepressants medicine to decrease depression presentation in exposed subjects to EMF.

Key words: Basil, depression, electromagnetic field (EMF), forced swimming test (FST).

INTRODUCTION

As recent increase in the use of electromagnetic field producing equipments, such as mobile phones, both epidemiological and experimental studies have been motivated. Indisputable reports from harmful effects of these microwaves have been associated with growing concern and some alarms in our today society. Exposure to electromagnetic field (EMF) at even low frequencies (900 to 1800 Hz) causes some established pathologic consequences such as increased permeability of the blood-brain barrier, disturbed neurons function and alteration in electroencephalography (EEG), disturbed regional cerebral blood flow, oxidant and antioxidant imbalance, neurotransmitter imbalance and genomic responses (Croft et al., 2002; Hamblin and Wood, 2002).

Cellular oxidation and free oxygen radicals release has been introduced as possible cellular injury mechanism that is accompanied with cognitional and affective sequences (Gülçin et al., 2007). In the following hypotheses, numerical antioxidative substances have been introduced to protect from central nervous system in front of oxidative effect of EMF. It is in infancy, but proceeds to protective agents in front of increasing environmental hazard factors, especially natural base agents have been motivated. There is more motivation to study herbal extracts as antioxidative agents, because herbal-base medications are accompanied with lower imposed side effects and have more facing today society (Katalinic et al., 2006). *Ocimum basilicum* (belongs to Labiateae family), have been employed traditionally in Iranian and Indian medicine as folklore remedy for a wide spectrum of ailments, also incorporated into a number of herbal medicinal preparations. It has been used as treatment of cold and persistent coughs. Some investigations

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shown its various protective effects including radiation protective efficacy, preventive potential against some chemicals, antiinflammatory effect, stimulant agent in central nervous system, bactericidal activity, modulatory effect on glutathione and improvement in cognitive task, antioxidant property, ulcer protective, antidiarrheal and blood-sugar (BS) lowering efficacy (Uma et al., 2000; Chattopadhyay, 1999). Basil safety in animal and human models has been confirmed (Katalinic et al., 2006). Recent study by Khaki et al. (2011) showed that basil could increase sperm health parameters and protect exposed animals by EMF. The objective of this research was to see the moderate effect of sweet basil on depression that is induced by EMF.

MATERIALS AND METHODS

Animals

Albino male Wistar rats, 8 weeks of age (weight = 220 ± 10 g) were housed in standard cages (14" × 9" × 8") inside a well-ventilated room, kept at $20 \pm 2^\circ\text{C}$ with 12-h dark-light cycle. They were obtained from Tabriz Medical Science University Central Animal Facility, and were kept in Dr. Khaki laboratory. All animals had free access to a standard pellet diet and water. Animals were exposed to radiation from experimental field (0.1 Tesla) for 6 h continuously per day for 6 weeks. Animals were free to move about in the cage during the exposure period. Sufficient ventilation and avoidance to impose heat shock to rats was controlled. Rats were brought back to the home cages following exposure. All interventions have been done during constant time period of 8:00 to 13:00.

Animals were divided into three groups: group I (n = 10) control group (no exposure to EMF without basil feeding), group II (n = 10) (exposure to EMF without basil feeding) and group III (n = 10) (exposure to EMF with basil feeding). All the experiments were performed in accordance with the European Animal Ethics Committee.

Forced swimming test

The modified forced swimming test was designed and used in accordance with the procedures described (Detke et al., 1997). Rats were taken and placed individually in a cylinder (50 × 20 cm) filled to a depth of 30 cm, by fresh, non-choleric water ($22 \pm 2^\circ\text{C}$). During the 5 min, the climbing, swimming and immobility behaviors were recorded at 5 s intervals.

Two swim sessions were conducted: a 15 min pretest, followed 24 h later by a 5 min test. The 5 min test sessions were videotaped and viewed at a later time by two raters blind to treatment. The raters have scored rat behavior for each 5 s period (60 times for the 5 min test) as one of the following: (1) immobility, making only those movements necessary to keep its head above water; (2) swimming, making active swimming movements; and (3) climbing, making vigorous movements with the forepaws in and out of the water, usually against the cylinder.

Tests were done in the second week and were repeated at the end of the study at the sixth week to evaluate acute and chronic effect of extract of basil.

Extract preparation

Fresh basil was prepared from local shopping in Tabriz.

Superfluous materials were rubbed off and were drained. Dried plants were steeped in methanol (80%), then extract was exploited in vacuum condition. Prepared extract was dried and used in maximum two days. Extracts were kept in refrigerator before use in laboratory. All of extract preparation processes have been done in pharmacognosy laboratory of Tabriz Pharmacology Faculty of Medicine, Tabriz-Iran.

Drug administration

Prepared extract was dissolved in aqua's water. Exposed rats were fed with basil extract as 0.5 g/kg body weight by nasogastric tube. Feeding was done at least 1 h before exposure to electromagnetic field.

Sucrose preference test

We have added standard sucrose consumption preference test to our experiment to confirmed depression presentation after exposure to electromagnetic field.

For the sucrose preference testing, all animals were presented with two bottles in their home cages. One bottle contained normal tap water and the second one 10% sucrose solution. Consumption rates were recorded every hour over a 3 h period. Sucrose was offered 1 h after daily feeding. Sucrose intake was expressed in relation of the animal's body weight. (ml/kg). Test was carried as a base score before exposure in both groups and would be repeated at the second and sixth weeks.

Statistical analysis

The data were analyzed by SPSS software (release 17). The number of time points where climbing, swimming or immobility were scored over the 5 min experimental session were summed for each rat. Data were expressed as means with standard error of mean (SEM). The means of measures in two groups were compared by independent-T test. Analyses of detected scores in behavioral parameters were done by one-way ANOVA test followed by Tukey's highest significant difference (HSD) test. P-value < 0.05 was considered as significant.

RESULTS

In the second week of the study, it was found that EMF exposure affected all parameters of forced swimming test, as it significantly increased immobility and decreased locomotor function of swimming and climbing in comparison to control group ($P < 0.05$). Basil feeding was not significantly effective to be protective in front of radiation imposed stress (Figure 1). In the last week, it was found that basil extract could be effective in decreasing immobility ($P < 0.001$) and in other way, in increasing swimming ($P < 0.001$). However, basil extract had not affected the climbing behavior significantly ($P = 0.18$) (Figure 2).

The Tukey's multiple comparisons test showed that basil extract in chronic stress (end of 6 weeks) significantly had decreased the total counts of immobility when compared with control ($P < 0.01$) and exposed group without basil feeding ($P < 0.01$). Basil extract had

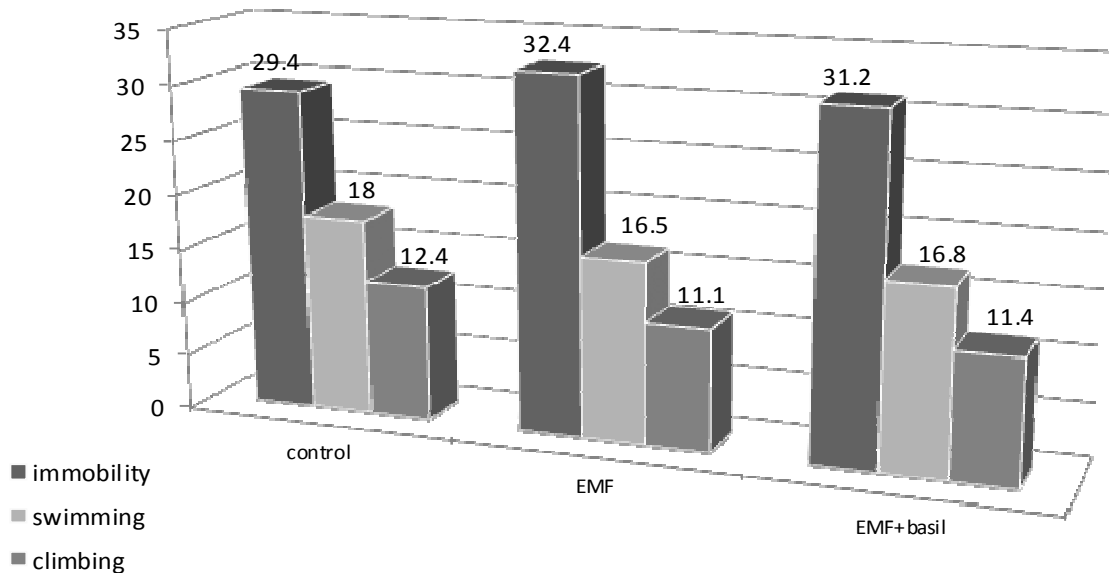


Figure 1. Effect of 1.5 g/kg body weight *O. basilicum* and 50 Hz EMF on forced swimming test parameters in the 2nd week.

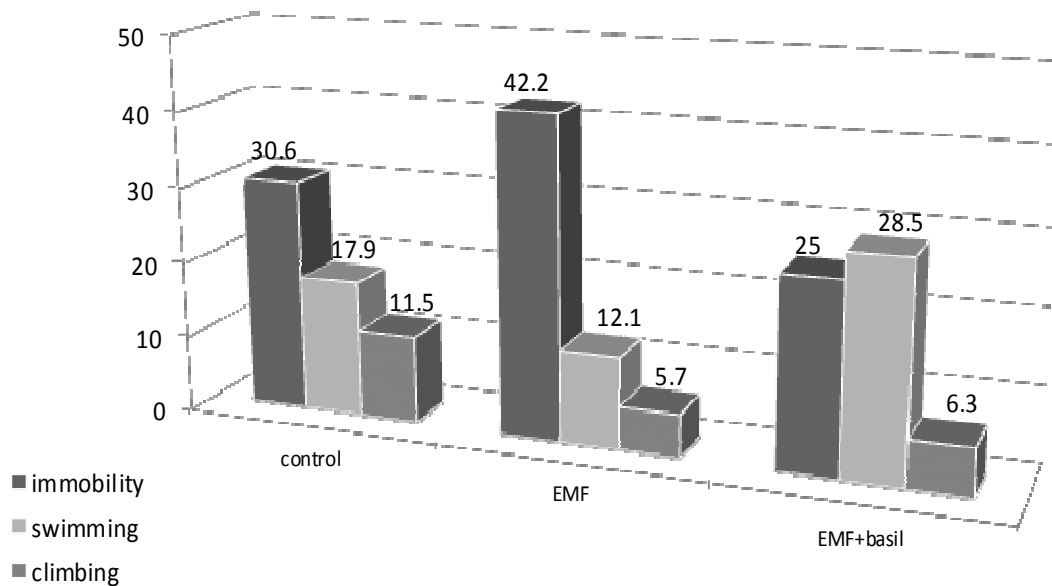


Figure 2. Effect of 1.5 g/kg body weight *O. basilicum* and 50 Hz EMF on forced swimming test parameters in the end of 6th week.

when compared with control ($P < 0.01$) and exposed group without extract ($P < 0.01$). Exposed animals had lower sucrose consumption in comparison to control group (29.5 ± 1.5 to 37.1 ± 1.1 , $P < 0.001$).

DISCUSSION

Animal models are indispensable tools in the search to

identify new antidepressant drugs (Urigen et al., 2008; Cryan et al., 2002). The forced swimming test (FST) is the most widely used animal test predictive of antidepressant treatment that was proposed primarily (Porsolt et al., 1977, 1978) and was subsequently improved (Detke et al., 1997; Cryan et al., 2005). The test is based upon the observation that rodents eventually develop immobility when they are placed in cylinder of water after they stop active escape behaviors, such as

increased the score of swimming behavior significantly climbing or swimming. Antidepressant treatments reduce the amount of immobility, or delay its onset, and increase or prolong active escape behaviors displayed during the FST. Our investigation showed that electromagnetic field exposure causes impose stress on animals that are followed by depression like behavior in exposed animals. The FST is the most widely used pharmacological model for assessment of potential antidepressant activity in rodents. Increased immobility factor in forced swimming test is indicator for depression like condition in animal. However, to exclude stress imposed by FST by self, we have carried out another standard test to evaluate depression in exposed animals. Sucrose preference test is a standard test that confirmed our hypothesis. Decreased sucrose consumption preference predicates anhedonia, the major component of depression in animal. It has been demonstrated that swimming score is sensitive to serotonergic compounds, and that climbing is sensitive to drug with selective effects on noradrenergic transmission (Page et al., 1999). Increasing swimming without significant increase in climbing like serotonergic agents predicts sedative effect of basil without stimulatory effect on locomotor activity. However, true mechanism of antidepressant effect of the *O. basilicum* is unknown but behavioral parameters in forced swimming test confirmed potential antidepressant effect as serotonergic agents. Unlike serotonergic pharmaceutical medicines, basil extract have not got serious side effects as sexual dysfunction or restlessness since our study have manifested that basil extract has antianxiety effects. Khaki et al. (2011) have shown that nourished animals with basil extracts had showed elevated level of testosterone and increased sexual desire as more sexual intercourse episodes.

Mood disorders have been treated with botanical medicines since antiquity, while modern research on herbal medicine in psychiatry, although, still in its infancy, has increased in recent years with a 50% increase in the literature (Garcia-Garcia et al., 2008). Several herbal medicine revealed on array of pre-clinical antidepressant activity. Some antidepressant herbal medicine, such as *Hypericum perforatum*, *Rhodiola rosea* (roseroot) and *Crocus sativas* (Saffron) offer promise for the treatment of depression via known psychopharmacological actions and sometimes antidepressant mechanisms are not as clearly defined as with selective serotonin reuptake inhibitors (SSRIs) (Toda and Morimoto, 2008; Panossian, et al., 2008; Limpeanchob et al., 2008; Sarris and Kvanagh, 2009; Van Dierman et al., 2009; Hosseinzadeh and Noraei, 2009; Krishnakumar et al., 2009; Awad et al., 2009; Lopez et al., 2009; Grundmann et al., 2009; Sena et al., 2009; Cao et al., 2010; Cho et al., 2010; Chang and Wang, 2010). Some agents, such as *Piper methysticum* and *H. perforatum* in meta-analyses studies revealed significant effects over placebo and comparable effects to synthetic agents (Linde, 2008; Sarris et al.,

2010). Herbal agents have better tolerability over some conventional antidepressants (Kasper et al., 2008a; 2010b). Although, these results and findings are encouraging, but there is so many non-tried herbal agents in traditional medicine of nations. *H. perforatum* is one of the mentioned herbal medicines that have is known as antidepressant herbal medicine since several years ago and its efficacy was evaluated by several experimental studies (Rojecky et al., 2004). Ishola et al. (2008) investigated antistress efficacy of extract of *Alchornea cordifolia* in rats by occupying forced swimming test.

Hosseinzadeh et al. (2007) investigated antidepressant effect of Kaemperol, a constituent of Saffron (*Crocus sativus*) on rats. Entirely, the present study has showed that *O. basilicum* extracts have had protective effect in exposed animals to electromagnetic fields induced depression. We suppose, protective property of basil against EMF is referred to its antioxidative potency and free radical scavenging activity. Some pervious studies have investigated basil antioxidative property in vital organs (Gülçin et al., 2007; Phuong et al., 2008). The antioxidative effect is mainly due to phenolic components, such as flavonoids, phenolic acids and phenolic diterpenes. The antioxidant activity of phenolic compounds is mainly due to their redox properties, which can play an important role in absorbing and neutralizing free radicals, quenching singlet and triplet oxygen, or decomposing peroxides (Shahidi et al., 1992). Comparing the mentioned studies with our study, showed similar results in their efficacy; nonetheless, basil is a nonexpansive material in general population with less probably hazard side effects that people have used from several and several hundreds years ago and it is acceptable in folklore culture with perfect favor in food. Fixed dose of feeding extract and the missing dose relative effect of basil on locomotor activity, and a synchronic comparison of different types of known antidepressants and the missing assessment of the biochemistry effect of basil on vital organs are the limitations observed in this study. We have tried to study a cost effective and easy obtained herbal substance to evaluate its protective efficacy in front of electro magnetic fields that enveloped us in every day life. We suggest more study on known herbal medicine drugs with more details in that field. We can not stop expansion of technology associated with EMF, but we can protect ourselves, especially young persons, against hazardous effects of radiations.

Conclusion

Basil extracts acted as antidepressant agents (by decreasing anhedonia and increasing locomotors) in vital subjects without or at least with lower side effects when compared to some known pharmaceutical agents, and were more pleasurable to patients.

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