Brief Report

Extremely low-frequency electromagnetic field influences the survival and proliferation effect of human adipose derived stem cells

Shahnaz Razavi, Marzieh Salimi[1], Daryoush Shahbazi-Gahrouei[1], Saeed Karbasi[1], Saeed Kermani[1]

Departments of Anatomical Sciences and [1]Medical Physics and Medical Engineering, School of Medicine, Isfahan University of Medical Sciences, Isfahan, Iran

Abstract

Background: Extremely low-frequency electromagnetic fields (ELF-EMF) can effect on biological systems and alters some cell functions like proliferation rate. Therefore, we aimed to attempt the evaluation effect of ELF-EMF on the growth of human adipose derived stem cells (hADSCs).

Materials and Methods: ELF-EMF was generated by a system including autotransformer, multi-meter, solenoid coils, teslameter and its probe. We assessed the effect of ELF-EMF with intensity of 0.5 and 1 mT and power line frequency 50 Hz on the survival of hADSCs for 20 and 40 min/day for 7 days by MTT assay. One-way analysis of variance was used to assessment the significant differences in groups.

Results: ELF-EMF has maximum effect with intensity of 1 mT for 20 min/day on proliferation of hADSCs. The survival and proliferation effect (PE) in all exposure groups were significantly higher than that in sham groups ($P < 0.05$) except in group of 1 mT and 40 min/day.

Conclusion: Our results show that between 0.5 m and 1 mT ELF-EMF could be enhances survival and PE of hADSCs conserving the duration of exposure.

Key Words: Extremely low-frequency electromagnetic fields, human adipose derived stem cells, proliferation effect, survival

Address for correspondence:
Prof. Shahnaz Razavi, Department of Anatomical Sciences, School of Medicine, Isfahan University of Medical Sciences, Isfahan, 81744-176, Iran. E-mail: razavi@med.mui.ac.ir
Prof. Daryoush Shahbazi-Gahrouei, Department of Medical Physics and Medical Engineering, School of Medicine, Isfahan University of Medical Sciences, Isfahan, 81744-176, Iran. E-mail: shahbazi@med.mui.ac.ir
Received: 05.02.2013, **Accepted:** 05.05.2013

Access this article online	
Quick Response Code:	Website: www.advbiores.net
	DOI: 10.4103/2277-9175.124668

INTRODUCTION

A primary epidemiological study showed an increase risk of leukemia in children who lived close to power line.[1] Other surveys at the 1st year indicated that extremely low-frequency electromagnetic fields (ELF-EMF) exposure can increase the risk of several cancers such as brain and breast tumors.[2,3] There is also a great concern in a possible association with breast cancer and electric power generation and consumption

How to cite this article: Razavi S, Salimi M, Shahbazi-Gahrouei D, Karbasi S, Kermani S. Extremely low-frequency electromagnetic field influences the survival and proliferation effect of human adipose derived stem cells. Adv Biomed Res 2014;3:25

in industrialized societies because breast cancer risk is considerably higher in developed countries than in other areas.

ELF-EMF, mostly emanating from electrical devices and transmission electric power lines with frequency of between 50 Hz and 60 Hz, surround our world now-a-days;[4] so it is important to know what is the effect of these exposure on human health. The effects of non-ionizing ELF-EMF chronic exposure on human beings has become a focus of interest since many years ago.[5,6] There is no reason that we assumed exposure to ELF-EMF is safe to our health; however, no causal relationship could be found until now between exposure to ELF-EMF and cancers and other abnormalities.[7,8]

In many research found that ELF-EMF can effect on biological systems and interferes with many biological functions in cells such as proliferation,[9] morphology,[10] apoptosis,[11] gene expression[12] and cell differentiation.[13] However, the mechanism of this effect is unknown yet.[14]

Results of bioelectro magnetics studies are contrary because of difference in the magnetic field intensity, frequency and duration of exposure.[7]

Even cell lines of different tissue origin and cell age may have diverse response to ELF-EMF exposure.[15] Hence, we need a standard "cellular model" for these studies to investigate better the effect of ELF-EMF on human beings.

Stem cells (SCs) are undifferentiated cells with the high renewable capacity that can differentiate to many other cell lineages.[16] High differentiation potentials of human mesenchymal stem cells (hMSCs) suggested that any changes in proliferation and metabolism of hMSCs may have unexpected results.[17] Therefore, it seems that the hMSCs can be appropriate models for assess the outcomes of ELF-EMF on biological systems.[18] Indeed; MSCs can be applied as the reference cells.

hMSCs isolated from some tissues such as bone marrow, umbilical cord blood and adipose tissue.[19] Adipose tissue has been known source of multi-potent stromal MSCs, which can be obtained by a less invasive method and in large amounts compared with bone marrow derived stem cells (BMSCs),[16] so in this study we used human adipose derived stem cells (hADSC).

Saino *et al*. showed that 2 mT and 75 Hz ELF-EMF increased the hMSCs proliferation rate.[20] It has been detected that human oral keratinocytes and rat glioma cells proliferation and differentiation was regulated by ELF-EMF.[21,22] The results of another study showed that ELF-EMF exposure on human neuroblastoma and rat pituitary cells increases the proliferation rate of these cells and inhibit programmed cell death.[11] It is found that exposure of ELF-EMF causes a reduction in proliferation and differentiation of hemopoietic SCs in compared with unexposed bone marrow cells.[23]

In a parallel research ELF-EMF 1 mT and 50 Hz could be inhibit the growth and proliferation rate of hMSCs, but had few influence on osteogenic differentiation of hMSCs.[18]

To consider the result of studies on the effect of on proliferation and differentiation was controversial and there is no evidence effect of ELF-EMF on hADSCs yet. Therefore, the result of ELF-ELF with intensity of 0.5 and 1 mT and 50 Hz (power line frequency) on the survival and proliferation effect (PE) of hADSCs for 20 and 40 min/day for 7 days was assessed.

MATERIALS AND METHODS

Magnetic field exposure system

A continuous sinusoidal 50 Hz magnetic field was generated by solenoid coils. The solenoid was wound with 720 turns of 1 mm enamel copper wire on a cylindrical core of acrylic tube (inner diameter: 20 cm, height: 24 cm). The solenoid was connected to an autotransformer, with a voltage percent scale, serially. Auto transformer was connected to 220 V power. The sinusoidal shape of signals to solenoid was evaluated by an oscilloscope connected to the solenoid. By setting the voltage percentage scale of auto transformer, the favorite flux density of magnetic field was obtained. The current and voltage to solenoid for each flux density were assessed by a digital multi-meter (digital HiTESTER.3256-50, Japan) connected to solenoid. Calibration of the system and uniformity was performed by a tesla-meter (LEYBOLD DIDACTIC GMBH 51662, Germany) with a probe AXIALE B-SONDE (model: 516.61). The uniformity of the EMF at the center of solenoid was ±1% where the cultures were located [Figure 1].

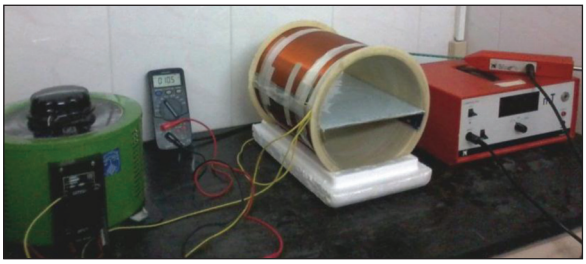

Figure 1: Extremely low-frequency electromagnetic fields generated system contained (from left to right) autotransformer, multimeter, solenoids coils, teslameter and its probe

The temperature in solenoid during the ELF-EMF exposure was controlled by a digital thermometer (Digital Hygro-Thermometer, France) with a probe that placed inside the solenoid indicate that magnetic field-induced heating was ignorable.

Isolation and culture of hADSCs

All procedures were conducted according to Isfahan University of Medical Sciences, Medical Faculty Ethic Committee approval. Human adipose tissue was obtained from three elective lipoaspirate samples of abdominal fat from female donors (age range: 23-41 year sold) after receiving informed consent. Briefly, samples were washed with sterile phosphate-buffered saline (PBS) to eliminate contaminating debris and red blood cells. Washed samples were treated with 0.075% collagenase type I in PBS for 30 min at 37°C with intermittent shaking. Then, the collagenase I was neutralized with an equal volume of DMEM/10% fetal bovine serum (FBS) and was centrifuged for 10 min at 750 rpm. The cellular pellet was resuspended in Dulbecco's Modified Eagle's Medium (DMEM), 10% FBS and plated in T25 flasks in 5 ml DMEM medium supplemented with 10% FBS and 1% Penicillin/streptomycin. After 24 h, the non-adherent cells were discarded and adherent cells were washed twice with PBS. When the confluency of cells was reached to 80-90% in the flask, the cells were passaged. After two passages, hADSCs were plated in 96-well plates and used to experiment. It is determined these isolated cells are stem in the previous study.[24] The cell cultures were kept at a temperature of 37°C in a humid atmosphere with 5% CO_2. The medium was changed every 3 days. All chemicals, except where specified otherwise, were purchased from Sigma — Aldrich, St. Louis, MO, USA.

ELF-EMF exposure

hADSCs were cultured to the 96-well plate at the density 10^3 cell/well and incubated for an overnight. Then, the plates were exposed to the ELF-EMF with intensities of 0.5 and 1 mT for 20 and 40 min/day for 7 days. The exposed cells were in four groups with the different EMF doses. Group 1: 1 mT for 40 min/day, group 2: 1 mT for 20 min/day, group 3: 0.5 mT for 40 min/day and group 4: 0.5 mT for 20 min/day. The sham groups were also placed in the turned off solenoid coils for 20 and 40 min/day. Conditions for exposure and sham groups were the same. In all experiments, the plates were located at the center of solenoid where the magnetic field was most uniform. Because of location of cell plates in out of incubator during the ELF-EMF exposure and control of this situation, one group of cells was placed in the incubator throughout all experiments time as the control group. Each test was performed for three times.

MTT assay

MTT (3-[4, 5-dimethylthiazol-2, 5-diphenyl tetrazolium bromid) assay was carried out for assessment the cell survival.

MTT was dissolved in PBS at 5 mg/ml. The stock solution was added to the culture medium at a dilution 1:10. The cell cultures were incubated at 37°C for 4 h. Then medium was aspirated and 100 µl of DMSO was added to extract the MTT formazan and the absorbance of each well was detected. To estimate the density of cells, MTT standard curve is depicted. The number of 50, 100, 500, 1000, 5000 and 10,000 hADSCs/well were incubated in 96 well plates. 10 µl MTT solutions was added to each well and the plates were incubated at 37°C for 4 h. After that the medium was aspirated and 100 µl of DMSO was added to each well. The absorbance of each well (each cell density) was detected by micro plate reader at the wavelength of 540 nm.

Proliferation effect assessment

Means and standard errors of the optical density (OD) of six replicates were calculated. The PE at each ELF-EMF dose was determined:[25] PE= average OD exposure group/average OD of the sham group.

Statistical analysis

One-way analysis of variance was used to assessment the significant differences in groups. Differences were significant when $^*P < 0.05$. All statistical analysis was performed with the Statistical Package for the Social Studies (SPSS 19, Chicago, IL, USA).

RESULTS

Morphology of hADSCs exposed to ELF-EMF

hADSCs with the confluency of 80% were modified to a spindle-shaped and fibroblastic morphology [Figure 2].[17] The morphological assessments by phase contrast microscope show that hADSCs incubated in 24 well plates exposed to 1 mT were oriented together to a common vector compared with hADSCs incubate in 96 well plates exposed to 1 mT [Figure 3].

Survival assay

The final number of cells that obtained from the MTT standard curve estimated as survival of hADSCs. The figure shows that the MTT standard curve line with y and r was linear ($y = 19775\ x - 2966.4$, $r = 9957$) [Figure 4].

The survival of hADSCs in all groups was significantly higher than one in sham groups except group of 1 mT, 40 min/day. This result showed that exposure of hADSCs with ELF-EMF not only is not toxic for the cells, but also increase the cell number [Figure 5].

Figure 2: Human adipose derived stem cells culture with confluence of 80% assessed by inverted microscope

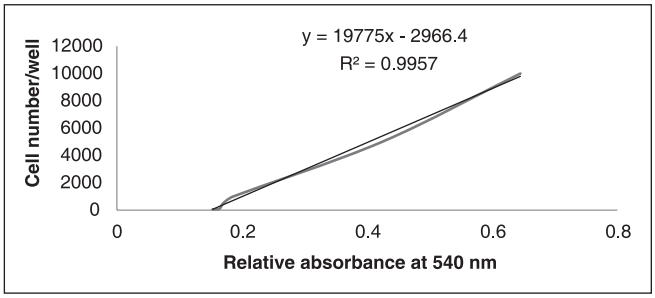

Figure 4: The MTT standard curve that plotted to estimate the cell number for optical density obtained from MTT assay

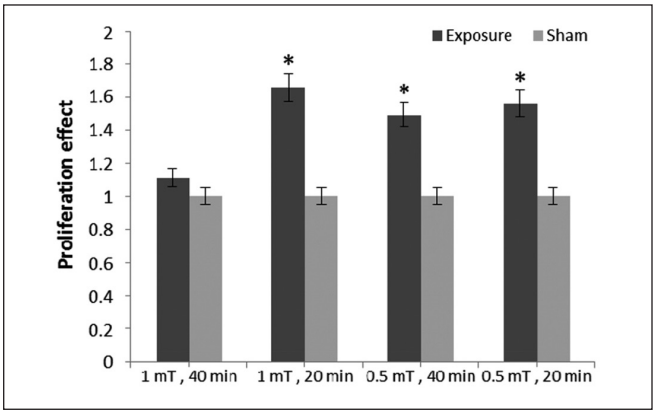

Figure 6: The effect of extremely low-frequency electromagnetic fields on proliferation effect PE) of human adipose derived stem cells after 7 days. The PE in all exposure groups was significantly higher than that in sham groups except in group of 1 mT, 40 min/day (**P* < 0.05). The PE of the incubated control group was no prominent difference with the sham groups

Proliferation effect

The PE in all exposure groups significantly was higher than that in sham groups except in group of 1 mT, 40 min/day (**P* < 0.05) [Figure 6].

DISCUSSION

The results of the current study show that short duration exposure of ELF-EMF can be increase the survival and PE of hADSCs significantly.

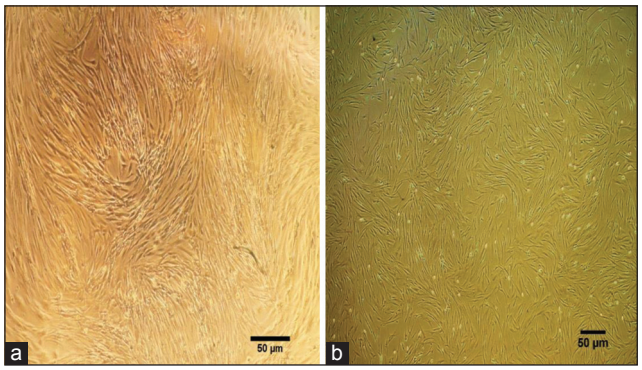

Figure 3: Morphology and orientation of cells exposed to extremely low-frequency electromagnetic fields in cultures in wells with different size. The cells in larger size of wells were oriented to an identified vector. (a) Human adipose derived stem cells (hADSCs) in 24 well plates. (b) hADSCs in 96 well plates

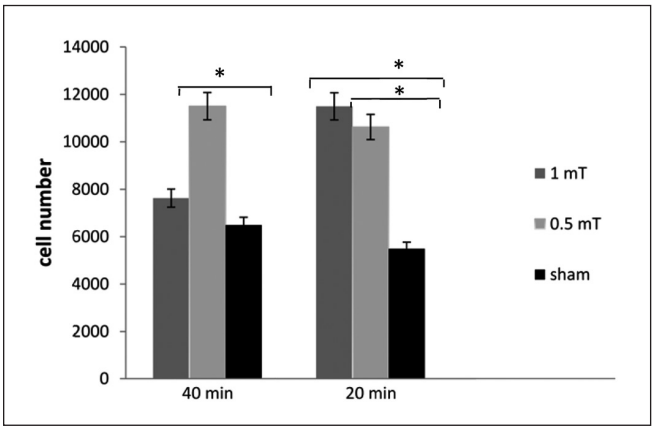

Figure 5: The effect of extremely low-frequency electromagnetic fields on survival of human adipose derived stem cells after 7 days. The survival in all exposure groups was significantly higher than that in sham groups except in group of 1 mT, 40 min/day (**P* < 0.05). In 40 min/day exposure, the survival in 0.5 mT intensity is significantly more than one in intensity of 1 mT (**P* < 0.05)

In this study, two intensities (0.5 and 1 mT) and duration (20 and 40 min/day) of ELF-EMF were selected to evaluate the effect of different intensities and duration of magnetic field on the survival and PE of hADSCs.

To assess the survival and PE of hADSCs MTT assay was applied. The survival and PE in exposure groups were more than ones in sham groups except in group of 1 mT and 40 min/day.

PE assessments show that ELF-EMF has maximum effect in the dose of 1 mT, 20 min/day. In all exposure groups PE was significantly more than sham groups except the group of 1 mT, 40 min/day.

In this study, we attempted to assess the effect of duration (20, 40 min/day) and intensity (0.5, 1 mT) on the survival and PE. It is no significant differences between groups of 0.5 mT with 20 and 40 min/day

exposure of ELF-EMF, but the survival of cells in 1 mT at 40 min group was less than that in group of 20 min/day exposure. With decrease the duration of exposure and radiation dose, the survival is increased in 1 mT intensity, so the survival of hADSCs has no direct association with exposure doses. In 40 min/day exposures, the cell survival in group of 0.5 mT is significantly more than one in group of 1 mT. In 20 min/day ELF-EMF exposure, no prominant difference was indicated between the hADSCs survival in 1 mT and 0.5 mT. It is probable that cells have similar response to different intensities of ELF-EMFs in short exposure durations; actually this is a quality response. The PE in all exposure groups was significantly higher than that in sham groups except in group of 1 mT, 40 min/day.

Because of location of solenoid coils outside of the incubator, it is possible that decrease in the survival of cells in group of 1 mT, 40 min is related to more duration that cells were in outside of the incubator. For assessment of this hypothesis, we used "control group in the incubator" that maintained in the incubator for all duration of experiment. The survival in this group was less than that in sham groups.

It is shown that the response of biological systems to ELF-EMF is the most intensive in the special intensities, durations and frequencies of electromagnetic fields (window effect). Indeed, the window effect is the window that biological systems show the most sensitivity to exposure of ELF-EMF.[26]

Exposure dose of ELF-EMF is the function of exposure duration and intensity of magnetic field. We used two intensities (0.5, 1 mT) and durations (20, 40 min/day) to assess the effect of different dosage of ELF-EMF.

Piacentini *et al*. showed that 1 mT, 50 Hz ELF-EMF for 6-24 h cause an increase in neural SCs proliferation. In this study, at 6 h duration was the most effect on cells and with the increase of exposure duration the difference in proliferation rate of neural SCs between the exposure and control groups was decreased.[13] The results of this study demonstrated that the effect of ELF-EMF on cell proliferation in shorter duration is more than that in longer duration in some intensity of ELF-EMF (1 mT).

In another study, application of ELF-EMF 3 mT to 3.6 mT for 30 min causes an increase in osteoblast cell proliferation.[27] Saino *et al*. indicated that ELF-EMF 2 mT increase the proliferation of osteoblast cells. The frequency selected was 75 Hz in their study.[20]

In some study, application of ELF-EMF make an inhibitory effect on cell proliferation such as study of Yan *et al*. that exposure of 20 mT ELF-EMF for 12 h/day inhibited the proliferation rate of hMSCs derived from bone marrow.[18]

Alterations in cell morphology, size and orientation can reveal cellular responses to EMF.[28] In our experiment, the morphology of hADSCs cultured in 96 well plates did not alter under exposure of ELF-EMF, whereas the cells in 24 well plates oriented to one vector.

Induced current (eddy current) by ELF-EMF in culture medium was calculated as follow: $I = \sigma\pi frB$ (i: Induced current, σ: Conductivity [σ was supposed to 1 s/m], f: Frequency, r: Radius of dish, B: Magnetic flux density).[29] The induced current (eddy current) in culture medium becomes lower as the size of culture flasks become smaller in 24 well plates the size of plates are more than that in 96 well plates; therefore, the induced eddy current in 24 well plates is more than the ones in 96 well plates. It is possible that the more eddy current induced in 24 well-plates effect on orientation of cultured hADSCs.

It is seem that the orientation of solenoid and produced magnetic field are important in acquired results. Harland and Liburdy in their study showed that the electrical field (E) induced from perpendicular magnetic field (B) on the culture dish is more than that in parallel magnetic field on culture dish by 5-6 folds because of E field depends on the cross-sectional area of the culture media seen by the B field.[30]

Studies performed so far have not identified the fundamental mechanisms inducing cellular responses to ELF-EMF. Cell proliferation and survival are very complex process controlled by multiple cell signal transduction pathways. Maintaining the integrity of genetic information during the cell proliferation is fundamental for living systems.[28]

One of the hypotheses is increasing the activity; concentration and life time of free radicals by Magnetic fields can be affect the kinetics of chemical reactions. Biological free radicals are highly reactive molecules that have unpaired electrons called "reactive oxygen species (ROS)".[7]

The other hypothesis is that ELF-EMF biological effects are followed by changing in intracellular Ca^{2+} signaling and homeostasis. Indeed, ELF-EMF is thought to be able to stimulate the activity of Ca^{2+} channels on the cell membrane.[13] The Ca^{2+} influx through these channels plays a key role in expression of special genes affected on cell functions such as differentiation and proliferation.[31] Bekhite *et al*.

indicated the relationship between MF-mediated intracellular ROS generation and Ca^{2+} as second messengers in signal pathways leading to cardio myocytes differentiation.[32]

In a study the extracellular Na^{+}/K^{+} concentration and osmolality increased after ELF-EMF exposure too. The authors presumed that it can be the reason of inhibition in cell proliferation under ELF-EMF exposure.[18]

CONCLUSION

Overall, our results showed that 50 Hz, 0.5 and 1 mT magnetic fields can be promote the survival and PE of the hADSCs regarding the duration of exposure; although, the effective mechanisms in this process are still unknown. More studies for assessment the effect of magnetic fields with other intensities and duration on SCs are suggested.

ACKNOWLEDGMNT

The authors are grateful to Isfahan University of Medical Sciences. We also thank Dr. Fereshteh Haghighat for providing human adipose tissue.

REFERENCES

1. Wertheimer N, Leeper E. Electrical wiring configurations and childhood cancer. Am J Epidemiol 1979;109:273-84.
2. Kheifets LI, Afifi AA, Buffler PA, Zhang ZW. Occupational electric and magnetic field exposure and brain cancer: A meta-analysis. J Occup Environ Med 1995;37:1327-41.
3. Feychting M, Forssén U, Floderus B. Occupational and residential magnetic field exposure and leukemia and central nervous system tumors. Epidemiology 1997;8:384-9.
4. Mann K, Röschke J. Sleep under exposure to high-frequency electromagnetic fields. Sleep Med Rev 2004;8:95-107.
5. Ahmed Z, Wieraszko A. The mechanism of magnetic field-induced increase of excitability in hippocampal neurons. Brain Res 2008;1221:30-4.
6. Czyz J, Nikolova T, Schuderer J, Kuster N, Wobus AM. Non-thermal effects of power-line magnetic fields (50 Hz) on gene expression levels of pluripotent embryonic stem cells-the role of tumour suppressor p53. Mutat Res 2004;557:63-74.
7. Sarvestani AS, Abdolmaleki P, Mowla SJ, Ghanati F, Heshmati E, Tavasoli Z, *et al*. Static magnetic fields aggravate the effects of ionizing radiation on cell cycle progression in bone marrow stem cells. Micron 2010;41:101-4.
8. Verkasalo PK, Pukkala E, Hongisto MY, Valjus JE, Järvinen PJ, Heikkilä KV, *et al*. Risk of cancer in Finnish children living close to power lines. BMJ 1993;307:895-9.
9. Tsai MT, Chang WH, Chang K, Hou RJ, Wu TW. Pulsed electromagnetic fields affect osteoblast proliferation and differentiation in bone tissue engineering. Bioelectromagnetics 2007;28:519-28.
10. Noriega-Luna B, Sabanero M, Sosa M, Avila-Rodriguez M. Influence of pulsed magnetic fields on the morphology of bone cells in early stages of growth. Micron 2011;42:600-7.
11. Grassi C, D'Ascenzo M, Torsello A, Martinotti G, Wolf F, Cittadini A, *et al*. Effects of 50 Hz electromagnetic fields on voltage-gated Ca^{2+} channels and their role in modulation of neuroendocrine cell proliferation and death. Cell Calcium 2004;35:307-15.
12. Mayer-Wagner S, Passberger A, Sievers B, Aigner J, Summer B, Schiergens TS, *et al*. Effects of low frequency electromagnetic fields on the chondrogenic differentiation of human mesenchymal stem cells. Bioelectromagnetics 2011;32:283-90.
13. Piacentini R, Ripoli C, Mezzogori D, Azzena GB, Grassi C. Extremely low-frequency electromagnetic fields promote *in vitro* neurogenesis via upregulation of Ca(v)1-channel activity. J Cell Physiol 2008;215:129-39.
14. Zhou J, Ming LG, Ge BF, Wang JQ, Zhu RQ, Wei Z, *et al*. Effects of 50 Hz sinusoidal electromagnetic fields of different intensities on proliferation, differentiation and mineralization potentials of rat osteoblasts. Bone 2011;49:753-61.
15. Sul AR, Park SN, Suh H. Effects of sinusoidal electromagnetic field on structure and function of different kinds of cell lines. Yonsei Med J 2006;47:852-61.
16. Pittenger MF, Mackay AM, Beck SC, Jaiswal RK, Douglas R, Mosca JD, *et al*. Multilineage potential of adult human mesenchymal stem cells. Science 1999;284:143-7.
17. Huang T, He D, Kleiner G, Kuluz J. Neuron-like differentiation of adipose-derived stem cells from infant piglets *in vitro*. J Spinal Cord Med 2007;30 Suppl 1:S35-40.
18. Yan J, Dong L, Zhang B, Qi N. Effects of extremely low-frequency magnetic field on growth and differentiation of human mesenchymal stem cells. Electromagn Biol Med 2010;29:165-76.
19. Fraser JK, Wulur I, Alfonso Z, Hedrick MH. Fat tissue: An underappreciated source of stem cells for biotechnology. Trends Biotechnol 2006;24:150-4.
20. Saino E, Fassina L, Van Vlierberghe S, Avanzini MA, Dubruel P, Magenes G, *et al*. Effects of electromagnetic stimulation on osteogenic differentiation of human mesenchymal stromal cells seeded onto gelatin cryogel. Int J Immunopathol Pharmacol 2011;24:1-6.
21. Manni V, Lisi A, Rieti S, Serafino A, Ledda M, Giuliani L, *et al*. Low electromagnetic field (50 Hz) induces differentiation on primary human oral keratinocytes (HOK). Bioelectromagnetics 2004;25:118-26.
22. Sul AR, Park SN, Suh H. Effects of electromagnetic fields on structure and function of rat glioma cell line. Res J Microbiol 2006;1:124-35.
23. Van Den Heuvel R, Leppens H, Nêmethova G, Verschaeve L. Haemopoietic cell proliferation in murine bone marrow cells exposed to extreme low frequency (ELF) electromagnetic fields. Toxicol *In Vitro* 2001;15:351-5.
24. Razavi S, Mardani M, Kazemi M, Esfandiari E, Narimani M, Esmaeili A, *et al*. Effect of leukemia inhibitory factor on the myelinogenic ability of Schwann-like cells induced from human adipose-derived stem cells. Cell Mol Neurobiol 2013;33:283-9.
25. Girgert R, Schimming H, Körner W, Gründker C, Hanf V. Induction of tamoxifen resistance in breast cancer cells by ELF electromagnetic fields. Biochem Biophys Res Commun 2005;336:1144-9.
26. Ivancsits S, Pilger A, Diem E, Jahn O, Rüdiger HW. Cell type-specific genotoxic effects of intermittent extremely low-frequency electromagnetic fields. Mutat Res 2005;583:184-8.
27. Cuccurazzu B, Leone L, Podda MV, Piacentini R, Riccardi E, Ripoli C, *et al*. Exposure to extremely low-frequency (50 Hz) electromagnetic fields enhances adult hippocampal neurogenesis in C57BL/6 mice. Exp Neurol 2010;226:173-82.
28. Simkó M. Induction of cell activation processes by low frequency electromagnetic fields. ScientificWorldJournal 2004;4 Suppl 2:4-22.
29. Oda T, Koike T. Magnetic field exposure saves rat cerebellar granule neurons from apoptosis *in vitro*. NeurosciLett 2004;365:83-6.
30. Harland JD, Liburdy RP. Environmental magnetic fields inhibit the antiproliferative action of tamoxifen and melatonin in a human breast cancer cell line. Bioelectromagnetics 1997;18:555-62.
31. Orrenius S, Zhivotovsky B, Nicotera P. Regulation of cell death: The calcium-apoptosis link. Nat Rev Mol Cell Biol 2003;4:552-65.
32. Bekhite MM, Figulla HR, Sauer H, Wartenberg M. Static magnetic fields increase cardiomyocyte differentiation of Flk-1(+) cells derived from mouse embryonic stem cells via Ca(2+) influx and ROS production. Int J Cardiol 2012;14489:11-22.

Source of Support: Nil, **Conflict of Interest:** None declared.