doi 10.15171/jrb.2014.10

Open Access Original Article

Journal of Radiobiology

Journal of Radiobiology
Vol. 1, No. 2, September 2014, 31-37
http://www.jradiobiol.com

Nickan Research Institute

Effect of extremely low-frequency (50 Hz) field on proliferation rate of human adipose-derived mesenchymal stem cells

Daryoush Shahbazi-Gahrouei[1*], Shahnaz Razavi[2], Marzieh Salimi[1]

Abstract

Introduction: The effects of non-ionizing extremely low-frequency magnetic fields (ELF-EMF) chronic exposure on human beings due to its potential health hazards has become a focus of interest since many years ago. Adipose tissue has been known as a source of multipotent stromal stem cells (MSCs), which can be obtained by a less invasive method and in large amounts compared with bone marrow derived stem cells (BMSCs). This study aimed to consider the effect of ELF-EMF on proliferation and differentiation rates of adipose-derived stem cells (ADSCs).

Methods: The effect of ELF-EMF with intensity of 0.5 and 1 mT and 50 Hz on proliferation rates of hADSCs at 20 and 40 min/day for 7 days was assessed. MTT assay was used to determine the growth and metabolism of cells and Trypan blue test was also done for cell viability.

Results: The lively hMSCs counting after ELF-EMF exposure at 7th day by Trypan blue staining showed that the proliferation in all groups of exposure was higher than the sham groups. There was an inconsistence result of MTT assay Trypan blue in group of 1 mT and 40 min/day exposure. In contrast to results of MTT assay, the proliferation rate of hMSCs in this group assessed by Trypan blue staining was significantly more than the sham group ($P<0.05$)

Conclusion: The results showed that 0.5 and 1 mT magnetic field strengths can be used to the proliferation rates of the (human mesenchymal stem cells) hMSCs derived from adipose tissue regarding the duration of exposure, although the effective mechanisms in this process are still unknown.

Keywords: Non-ionizing radiation, Extremely low-frequency magnetic fields, Proliferation rate, Human multipotent stromal stem cells (hMSCs)

Introduction

Extremely low-frequency electromagnetic fields (ELF-EMF) are ubiquitous in our environment more than the past. Electrical devices are the good examples from which ELF-EMF is generated. The transmission power lines with frequency of 50 Hz serve as important sources of human exposure to these fields (1).

The effects of non-ionizing ELF-EMF chronic exposure on humans, due to its potential health hazards, has become the focus of interest since many years ago (1,2). Epidemiological studies on children who lived close to power line have shown an increased risk of leukemia (3). Other surveys suggested that ELF-EMF exposure can increase the risk malignancy in brain and breast (4,5). There is still not enough evidence to prove that the exposure to ELF-EMF is harmful for human health (6); and no relationship could be found until now, neither for cancers nor for other abnormalities (7,8).

Many research works over the last 20 years showed that ELF-EMF can have many effects on biological systems and disturb many biological functions in the cell such as proliferation (9,10), morphology (11), apoptosis (12), gene expression (13) and cell differentiation (14); although the mechanisms behind its effect are still unknown (6,15). Bioelectromagnetics studies have shown that differences in magnetic field intensity, frequency and duration of exposure can affect outcome (6). Cell lines of diverse tissue origin may also have different reaction to ELF-EMF (16). Among the first studies is the one from Liboff et al. that assessed the influence of sinusoidal field with intensity of 0.16 μT and 76 Hz on fibroblast proliferation. The authors reported a correlation between ELF-EMF and proliferation of fibroblast (17). De Mattei et al. has also indicated that the magnetic field exposure can increase the proliferation of TE-85 and MG-63 cells (18).

Stem cells (SCs) as undifferentiated cells have high renewable capacity and can differentiate to many other cell lineages (19,20). SCs can be applied as the reference cells (15,21,22). In addition, these cells are found in tumors such as brain and breast, therefore any effect of ELF-EMF on SCs may be effect on cancer tissues (23,24).

High differentiation potentials of human mesenchymal

Received: 3 August 2014 2014, **Accepted:** 17 August 2014, **Published online:** 1 September 2014

[1]Department of Medical Physics and Medical Engineering, School of Medicine, Isfahan University of Medical Sciences, Isfahan, Iran
[2]Department of Anatomical Sciences, School of Medicine, Isfahan University of Medical Sciences, Isfahan, Iran
***Corresponding Author:** Prof. Daryoush Shahbazi-Gahrouei, Professor of Medical Physics, Department of Medical Physics and Medical Engineering, Isfahan University of Medical Sciences, Isfahan, Iran. Phone: +98-311-7922495, Email: Shahbazi@med.mui.ac.ir

■ Implication for health policy/practice/research/ medical education

The effects of non-ionizing extremely low-frequency electromagnetic fields (ELF-EMF) chronic exposure on humans, due to its potential health hazards, has become the focus of interest since many years ago. Epidemiological studies on children who lived close to power line have shown an increased risk of leukemia. Other surveys suggested that ELF-EMF exposure can increase the risk of malignancy in brain and breast. However, there is still not enough evidence to prove that the exposure to ELF-EMF is harmful for human health, nevertheless, no relationship could be found until now, neither for cancers nor for other abnormalities.

stem cells (hMSCs) suggested that any changes in proliferation and metabolism of hMScs may have unexpected results. Therefore, it seems that the SCs can serve as an appropriate model to assess the outcomes of ELF-EMF on biological systems. Human mesenchymal stem cell (MSCs) is a kind of stem cells that have the potential for differentiation into adipocyte, chondrocyte and osteocyte (19). They are isolated from some tissues such as bone marrow, umbilical cord blood and adipose tissue (25).

Saino et al. showed that 2 mT and 75 Hz ELF-EMF increased the hMScs proliferation rate (26). It has been found that EMFs regulate proliferation and differentiation of human oral keratinocytes and rat glioma cells (16,27). The results of previous study showed that ELF-EMF exposure on human neuroblastoma and rat pituitary cells increased the proliferation rate of these cells and inhibit programmed cell death (12). While, the results of other study showed that exposure of ELF-EMF induced a reduction of proliferation and differentiation of haemopoietic cells in compared to unexposed bone marrow cells (28). In a similar research ELF-EMF 1 mT and 50 Hz could be inhibit the growth and proliferation rate of hMSCs but minor effect on osteogenic differentiation of hMSCs (15).

There is no enough evidence to consider the effect of ELF-EMF on human ADSCs and the study results are controversial. Therefore, this study aimed to consider the effect of ELF-EMF on proliferation and differentiation rates of hADSCs with intensity of 0.5 and 1 mT and 50 Hz (power line frequency) for 20 and 40 min/day and duration of 7 days.

Methods

Magnetic field exposure system

A continues sinusoidal 50 Hz magnetic field was generated by a solenoid coil. 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. Autotransformer 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 autotransformer, the favorite flux density of magnetic field was obtained. The current and voltage of solenoid for each flux density were assessed by a digital multimeter (digital HiTESTER.3256-50, Japan) connected to solenoid. The current and voltage for 0.5 mT were 202.1 mA and 4.68 V and for 1 mT were 9.19 V and 395 mA, respectively. Calibration of system and uniformity was performed by a teslameter (LEYBOLD DIDACTIC GMBH 51662, Germany) with a probe AXIALE B-SONDE (model: 516.61). The uniformity of the EMF at the center of the solenoid, where the cell cultures were located, was ±1%.

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

Isolation and culture of hMSCs

Adipose tissue has been known source of multipotent stromal MSCs, which can be obtained by a less invasive method and in large amounts compared with BMSCs (19), so here study was done on ADSCs (adipose-derived stem cells).

All procedures were conducted according to Isfahan University of Medical Sciences's ethical committee approval. Human adipose tissue was obtained from three elective lipoaspirate samples of abdominal fat from female donors (age range: 23–41 years old) after receiving informed consent. Briefly, samples were washed with sterile phosphate-buffered saline (PBS) to eliminate contaminating debris and red blood cells (RBC). The samples were then 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 then was centrifuged for 10 min at 750 rpm. The cellular pellet was resuspended in DMEM/10% FBS and plated in T25 flasks in 5 ml DMEM (F12 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. It is determined these isolated cells are indeed stem cells in the previous study (29). When the confluency of cells was reached to 80-90% in the flasks, the cells were passaged with Trypsin/EDTA harvesting (Figure 1). After two passages, hMSCs were plated in 96-well plates and used to treat with ELF-EMF. The cultures

were kept at a temperature of 37 °C in humid atmosphere with 5% CO2. The medium was changed every 3 days. All chemicals, except where specified otherwise, were purchased from Sigma–Aldrich, St. Louis, MO, USA.

Magnetic field exposure protocols

Human mesenchymal stem cells (hMSCs) were inoculated to the 96-well plate at the density 103 cell/well and were grown overnight. Then, the plates were exposed to the 50 Hz 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 including: 1 mT and 0.5 mT for both two times of 20 min/day and 40 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. All tests were performed for three times.

MTT proliferation assay

MTT (3-(4,5-dimethylthiazol- 2,5–diphenyl tetrazolium bromid) assay was carried out to assess the viability and proliferation rate of hMSCs. MTT was dissolved in PBS at 5 mg/ml. The stock solution was added to the culture medium at a dilution 1:10. The plates were incubated at 37 °C for 4 h. The medium was then aspirated and 100 μl of DMSO was added to extract the MTT formazan and the absorbance of each well was detected by microplate reader (Hiperion MPR 4+, Germany) at the wavelength of 540 nm. For estimating the number of cells, MTT standard cure was depicted.

Trypan blue test

The trypan blue dye exclusion assay was used for measurement of ADSCs viability. The medium in wells was aspirated. 0.1 ml of trypan blue 0.4% in PBS stock solution added to 0.1 ml of cells. The hemacytometer was loaded and examined immediately under a microscope at low magnification. The number of viable cells—small, round and refractive—was counted. The non-viable cells were swollen, larger and dark blue.

Growth kinetics

The doubling time was calculated to determine hMSCs proliferation potential. Groups of hMSCs in media supplemented with 10% FBS were plated in well at 1000 cells/cm2. After harvesting confluent cultures the number of cell in suspension were counted to calculate the doubling time (30). Doubling time was obtained using the following equation:

$$\text{Doubling Time} = T\,[\ln 2/\ln (N_2/N_1)]$$

where T is expansion days; N_1 and N_2 are the initial and final cell numbers.

Statistical analysis

In all graphs error bars (± s.e.m.) are shown for independent experiments. The results from different experiments were pooled and the number of independent data points (n) is indicated in the figure legends of the corresponding graphs. Significances were calculated using an ANOVA (one-way and two way as indicated) approach and were corrected by the Bonferroni post hoc test.

Results

Morphology of hMSCs exposed to ELF-EMF

Adipose–derived stem cells (ADSCs) with the confluency of 80% were modified to a spindle-shaped and fibroblastic morphology according to Huang et al. work (19) (Figure 1). The morphological assessments by phase contrastmicroscope did not show any prominent difference between exposed and control groups (Figure 2).

Cell Viability

To determine the hMSCs proliferation rate after ELF-EMF exposure at 7th day, MTT method was applied. The proliferation rate of hMSCs in exposure groups was significantly higher related to sham groups except in group of 1 mT and 40 min/day ($P<0.05$) (Figure 3). Calculation of doubling time confirmed high proliferation

Figure 1. hMSCs culture with confluence of 80% assessed by inverted microscope.

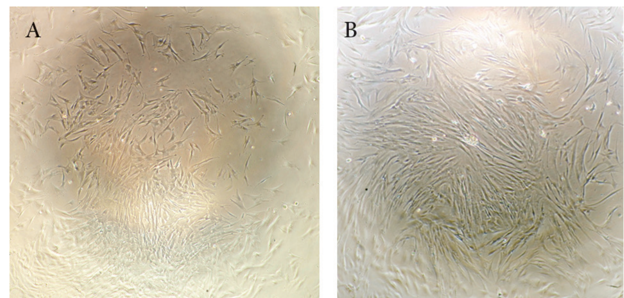

Figure 2. Morphology of exposure and sham groups. The morphological assessments did not indicate any significant difference between exposed (A) and control groups (B).

potency of hMSCs exposed to ELF-EMF compared to the control groups except in group which exposed with 1 mT, 40 min/day.

Proliferation rate of hMSCs assessed by Trypan blue staining

The lively hMSCs counting after ELF-EMF exposure at 7th day by Trypan blue staining show that the proliferation in all groups of exposure was higher than the sham groups. There is an inconsistence result of MTT assay Trypan blue in group of 1 mT, 40 min/day. In contrast to results of MTT assay, the proliferation rate of hMSCs in this group assessed by Trypan blue staining is more than the sham group significantly (P<0.05) (Figure 4). The doubling time in exposure groups was significantly lower than sham groups (*P<0.05) except in group of 1 mT, 40 min/day (Figure 5).

Discussion

The results of current study show that short duration exposure of ELF-EMF can increase the proliferation rates of hMSCs significantly. However, ELF-EMF has not enough energy to cause ionization of molecules in the cells as it is classified as non-ionizing radiation (1,11). The dose

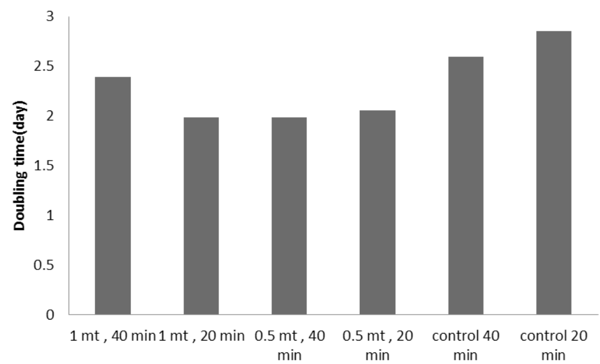

Figure 5. The effect of ELF-EMF on the doubling time of hMSCs. The doubling time in exposure groups was lower than sham groups significantly (*P<0.05) except in group of 1 mT, 40 min/day.

of radiation is a function of field strength and duration of exposure (31). In this study, two intensity (0.5 and 1 mT) and duration (20 and 40 min/day) of ELF-EMF was selected to evaluate the effect of different intensities and duration of magnetic field on proliferation rate of cells.

To assess the proliferation rate of hMSCs, MTT assay and Trypan blue staining were performed. In MTT assay the results showed an increased proliferation rate by MTT and trypan blue assay compared to sham groups. The doubling time calculations approved the MTT results, whereas in Trypan blue staining, all exposure groups had more proliferation than the sham groups.

In this study proliferation rate in cells treated with 0.5 mT and 1 mT for duration of 20 and 40 min/day was determined. Our results showed that low rate of proliferation in the cells exposed to 1mT for 40 min/day was less than the ones exposed to 1 mT for 20 min/day.

Our results also indicate that with a decrease in the duration of exposure and radiation dose, proliferation rate will be increased. So the proliferation rate of hMSCs has no direct association with exposure doses. Since the location of solenoid coils are outside the incubator, it is likely to think that the decrease in proliferation of cells in group of 1 mT, 40 min is because the cells were exposed to a lesser duration time comparing with the ones in the incubator. The proliferation in this group was less than in the others. The difference between groups of 1 mT 40 min and 0.5 mT, 40 min is significant and is shown by a decrease of ELF-EMF intensity from 1 mT to 0.5 mT the amount of proliferation of cells is increased. Consistent with that, result of Trypan blue staining showed that proliferation rate of hMSCs in all exposure groups is more than those in the sham groups.

The comparison of hMSCs proliferation rates among groups of 1 mT for 20 min and 40 min exposure with 0.5 mT for 20 min and 40 min exposure showed no significant differences. The proliferation in groups of 0.5 mT was more than that in the groups of 1 mT.

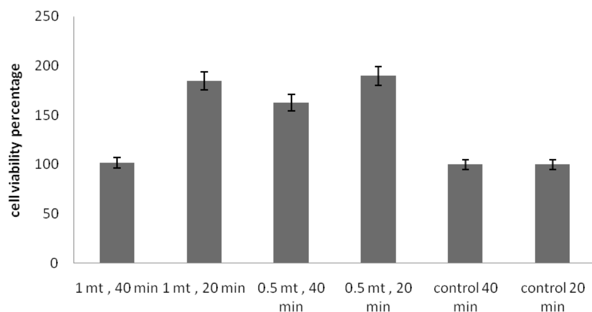

Figure 3. The effect of ELF-EMF on viability of hMSCs detected by MTT assay. The percent of viable cells in exposure groups was significantly higher than sham groups (*P<0.05), except in group of 1 mT, 40 min/day.

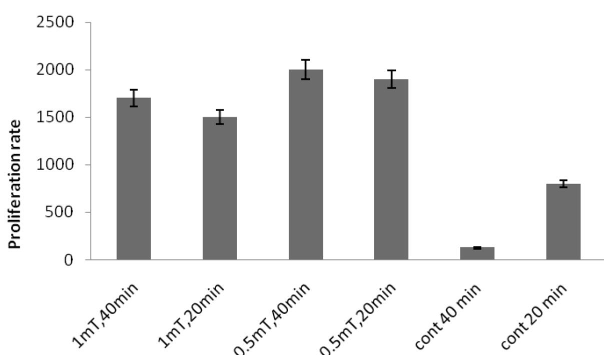

Figure 4. The effect of ELF-EMF on proliferation rate of hMSCs detected by Trypan blue. The proliferation rate in all exposure groups was higher than sham groups significantly (*P<0.05).

It is shown that the response of biological systems to ELF-EMF is significantly intensive in the special intensities, durations and frequencies of electromagnetic fields. Indeed, the window effect is the window that biological systems show the most sensitivity to exposure of ELF-EMF (32). Here, it is seems that window effect for this special cell line (hMSCs) and frequency (50 Hz) is in near 0.5 mT.

The results are in good agreement with those reported by Piacentini et al. who showed an increase in neural stem cells proliferation by applying a field of 1 mT, 50 Hz for 6-24 h (33). Recently, 6 h duration was the most effect on cells and with increase of exposure duration the difference in proliferation rate of NSCs between the exposure and control groups was decreased (33).

In a similar study, application of ELF-EMF 3 mT to 3.6 mT for 30 min causes an increase in osteoblast cell proliferation (34). In some other studies, application of ELF-EMF make an inhibitory effect on cells 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 (15).

Previous studies that prove the cellular responses are dependent on duration of ELF-EMF exposure is survey of Sul et al. who assessed the effect of 2 mT, 60 Hz on four cell lineage hFOB 1.19, T/G HA-VSMC cells, RPMI 7666 cells and HCN-2 for different duration of 1 h/day, 3 h/day and 6 h/day for 7 and 14 days (16). The results of this study showed that in each cell line, only one of the durations is the most effective one on cell proliferation. Although hFOB cells proliferation was elevated with increase in exposure duration, the other 3 linages showed the non-linear response to exposure of ELF-EMF. The mechanisms of this effect on biological systems are still elusive.

One of the hypotheses is that increasing the activity, concentration and life time of free radicals by magnetic fields can be affect the kinetics of chemical reactions with radical pair intermittent (6).

Piacentini et al. assumed that charged molecules like Salic acid on outer part of the cell could be affected by magnetic fields and regulate proliferation of cells (33). 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 (33,35). 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 (36). In a study the extracellular Na+/K+ concentration and osmolality increased after ELF-EMF exposure. The authors presumed that it can be the reason of inhibition in cell proliferation under ELF-EMF exposure (15).

Conclusion

Overall, the results showed that 50 Hz, 0.5 and 1 mT magnetic fields may be promoted the proliferation rates of the hMSCs derived adipose tissue 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 stem cells are suggested.

Acknowledgments

None declared.

Authors' contributions

All authors wrote the manuscript equally.

Ethical considerations

Ethical issues (including plagiarism, data fabrication, double publication) have been completely observed by the author.

Conflict of interests

The authors declared no competing interests.

Funding/Support

None declared.

References

1. Mann K, Roschke J. Sleep under exposure to high-frequency electromagnetic fields. Sleep Med Rev 2004; 8: 95-107. doi: 10.1016/S1087-0792(03)00004-2
2. 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 tumor suppressor p53. Mutat Res 2004; 557(1): 63-74. doi: 10.1016/j.mrgentox.2003.09.011
3. Wertheimer N, Leeper E. Electrical wiring configurations and childhood cancer. Am J Epidemiol 1979; 109: 273-84.
4. Feychting M. Health effects of static magnetic fields: a review of the epidemiological evidence. Prog Biophys Mol Biol 2005; 87: 241-6. doi: 10.1016/j.pbiomolbio.2004.08.007
5. Repacholi MH, Greenebaum B. Interaction of static and extremely low frequency electric and magnetic fields with living systems: health effects and research needs. Bioelectromagnetics 1999; 20: 133-60.
6. 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. doi: 10.1016/j.micron.2009.10.007
7. Schreiber GH, Swaen GM, Meijers JM, Slangen JJ, Sturmans F. Cancer mortality and residence near electricity transmission equipment: a retrospective cohort study. Int J Epidemiol 1993; 22: 9-15.

doi: 10.1093/ije/22.1.9
8. Tomenius L. 50 Hz electromagnetic environment and the incidence of childhood tumors in Stockholm County. Bioelectromagnetics 1986; 7: 191-207. doi: 10.1002/bem.2250070209
9. Schimmelpfeng J, Dertinger H. Action of a 50 Hz magnetic field on proliferation of cells in culture. Bioelectromagnetics 1997; 18: 177-183. doi: 10.1002/(SICI)1521-186X(1997)18:2<177::AID-BEM11>3.0.CO;2-O
10. 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(7): 519-28. doi: 10.1002/bem.20336
11. 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(6): 600-7. doi: 10.1016/j.micron.2011.02.005
12. Grassi C, D'Ascenzo M, Torsello A, Martinotti G, Wolf F, Cittadini A, et al. Effect 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. doi: 10.1016/j.ceca.2003.09.001
13. Mayer-Wanger S, Passberger A, Sievers B, Aigner J, Summer B. Effect of low frequency electromagnetic fields on the chondrogenic differentiation of human mesenchymal stem cell. Bioelectromagnetics 2011; 32(4): 283-90. doi: 10.1002/bem.20633
14. Lisi A, Ledda M, Rosola E, Pozzi D, Emilla ED, Giuliani L, et al. Extremely low frequency electromagnetic field exposure promotes differentiation of pituitary corticotrope-derived AtT20D16Vcells. Bioelectromagnetics 2006; 27: 641-51. doi: 10.1002/bem.20255
15. Yan J, Dong L, Zhang B, Qi N. Effects of extremely low-frequency magnetic field on growth and differentiation of human mesenchymal stem cells. J Electromagnetic Biol Med 2010; 29 (4): 165-76. doi: 10.3109/01676830.2010.505490
16. 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(6): 852-61. doi: 10.3349/ymj.2006.47.6.852
17. Liboff AR, Williams T Jr, Strong DM, Wistar R Jr. Time-varying magnetic fields: effect on DNA synthesis. Science 1984; 223: 818-20. doi: 10.1126/science.6695183
18. De Mattei M, Caruso A, Traina GC, Pezzetti F, Baroni T, Sollazzo V. Correlation between pulsed electromagnetic fields exposure time and cell proliferation increase in human osteosarcoma cell lines and human normal osteoblast cell in vitro. Bioelectromagnetics 1999; 20(3): 177-82.
19. 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(1): 35-40.
20. 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. doi: 10.1126/science.284.5411.143
21. Wobus AM, Guan K, Yang HT, Boheler KR. Embryonic stem cells as a model to study cardiac, skeletal muscle, and vascular smooth muscle cell differentiation. Methods Mol Biol 2002; 185: 127-56. doi: 10.1385/1-59259-241-4:127
22. Spielmann H, Pohl I, Doring B, Liebsch M, Moldenhauer F. The embryonic stem cell test, an in vitro embryotoxicity test using two permanent mouse cell lines: 3T3 fibroblasts and embryonic stem cells. In Vitro Toxicol 1997; 10: 119-27.
23. Ablett MP, Singh JK, Clarke RB. Stem cells in breast tumours: Are they ready for the clinic? Eur J Cancer 2012; 48: 2104-16. doi: 10.1016/j.ejca.2012.03.019
24. Singh SK, Clarke ID, Terasaki M, Bonn VE, Hawkins C, Squire J, et al. Identification of a Cancer Stem Cell in Human Brain Tumors. Cancer Res 2003; 63: 5821-8.
25. Fraser JK, Wulur I, Alfonso Z, Hedrick MH. Fat tissue: an underappreciated source of stem cells for biotechnology. Trends Biotechnol 2006; 24(4): 150-4. doi: 10.1016/j.tibtech.2006.01.010
26. Saino E, Fassina L, Van Vlierberghe S, Avanzini MA, Dubruel P, Magenes G, et al. Effect of electromagnetic stimulation on osteogenic differentiation of human mesenchymal stromal cells seeded onto gelaticryogel. Int J Immunopathol Pharmacol 2011; 24: 1-6.
27. Manni V, Lisi A, Rieti S, Serafino A, Ledda M, Giuliani L. Low electromagnetic field (50 Hz) induces differentiation on primary human oral keratinocytes (HOK). Bioelectromagnetics 2004; 25: 118-26. doi: 10.1002/bem.10158
28. Van den Heuvel R, Leppens H, Ne methova 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. doi: 10.1016/S0887-2333(01)00035-2
29. Razavi S1, 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(2): 283-9. doi: 10.1007/s10571-012-9895-2
30. Shafaei H, Esmaeili A, Mardan M, Razavi S, Hashemiben B. Effects of human placental serum on proliferation and morphology of human adipose tissue-derived stem cells. Bone Marrow Transplant 20114; 6: 1464-71. doi: 10.1038/bmt.2010.313
31. Chang K, Chang WH. The influence of pulsed electromagnetic field (PEMF) on osteopenia in an

ovariectomized female rat model. In: Proceedings of the 23rd Annual Meeting of the Bioelectromagnetics Society St Paul, MN 2001.

32. Ivancsits S, Pilger A, Diem E, Jahn O, Rudiger HW. Cell type specific genotoxic effects of intermittent extremely low-frequency electromagnetic fields. Mutat Res 2005; 583: 184-8. doi: 10.1016/j.mrgentox.2005.03.011
33. 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(1): 129-39. doi: 10.1002/jcp.21293
34. 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. doi: 10.1016/j.expneurol.2010.08.022
35. Bekhite MM, Figulla HR, Sauer H, Wartenberg M. Static magnetic fields increase cardiomyocyte differentiation of Flk-1cells derived from mouse embryonic stem cells via Ca2+influx and ROS production. Int J Cardiol 2013; 167(3): 798-808. doi: 10.1016/j.ijcard.2012.02.020
36. Orrenius S, Zhivotovsky B, Nicotera P. Regulation of cell death: the calcium apoptosis link. Nat Rev Mol Cell Biol 2003; 4: 552-65. doi: 10.1038/nrm1150

Please cite this paper as: Shahbazi-Gahrouei D, Razavi S, Salimi M. Effect of extremely low-frequency (50 Hz) field on proliferation rate of human adipose-derived mesenchymal stem cells. J Radiobiol 2014; 1(2): 31-37. doi: 10.15171/jrb.2014.10

Copyright © 2014 The Author(s); Published by Nickan Research Institute. This is an open-access article distributed under the terms of the Creative Commons Attribution License (http://creativecommons.org/licenses/by/4.0), which permits unrestricted use, distribution, and reproduction in any medium, provided the original work is properly cited.