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Effect of Faradarmani Consciousness Field on Proliferation, Telomerase Activity, and Telomere Length of Human Mesenchymal Stem Cells

Mohammad Ali Taheri[1], Laleh Amani[2], Simin Sharifi[3], Noushin Nabavi[4], Ali Zaman Vaziri[5], Ahmad Khalili[5*]

* Corresponding author: Ahmad Khalili
Email: Khalili.a.cbc@gmail.com

1. Sciencefact R&D Department, Cosmointel Inc. Research Center, Ontario, Canada
2. Department of Biology, Science and Research Branch, Islamic Azad University, Tehran, Iran
3. Department of Pharmaceutical Biotechnology, Faculty of Pharmacy, Tabriz University of Medical Sciences, Tabriz, Iran
4. Research services at the University of Victoria, Victoria, British Columbia, Canada
5. Cancer Biomedical Center (CBC) Research Institute, Tehran, Iran

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Abstract

Taheri Consciousness (T-Consciousness) was introduced and defined by Mohammad Ali Taheri as one of the constituent components of the Cosmos, in addition to matter and energy, from which Taheri Consciousness Fields (TCFs) derive. TCFs are not matter or energy, but scientific experiments can prove them. The effect of Faradarmani CF, as one of the TCFs, was examined in this study. Telomerase is an important enzyme that adds DNA sequence repeats to the ends of chromosomes, thus preventing their shortening. Telomerase activity is associated with cell proliferation, organism growth, and aging. This study aimed to study proliferation, telomerase activity, and telomere length in the Faradarmani CF treated human mesenchymal stem cells (hMSCs). Human MSCs were isolated from bone marrow, and the morphology, cell proliferation, telomere length, and telomerase activity were evaluated in the Faradarmani CF treatment and control groups. The results showed that Faradarmani CF significantly increased proliferation, telomere length, and telomerase activity in hMSCs compared with the control group ($p < 0.05$). Given the effects of Faradarmani CF on telomere length and telomerase activity, which are essential factors in the treatment of several age-related diseases, it is recommended that additional experiments be conducted to clarify the effects of TCFs on aging.

Keywords: Taheri Consciousness Fields, Faradarmani Consciousness Field, telomerase activity, telomere length, mesenchymal stem cells

Introduction

The discovery of telomere structure and the accurate description of their functions were important achievements in molecular medicine, especially in gerontology (Blackburn, 2001; Kim et al., 2002). To date, several experimentations and data from long-term medical investigations have confirmed the validity of the telomere theory of aging that associates life expectancy and the progress of age-related diseases with telomere length (Calado et al., 2012; Hoen et al., 2011; Martin-Ruiz et al., 2005; Zglinicki et al., 2005). A reverse association has been found between telomere length and various forms of cancer (Broberg et al., 2005; Bagheri et al., 2006), renal failure (Boxall et al., 2006; Ramírez et al., 2005), cardiovascular disease (Demissie et al., 2006; Zhan et al., 2019), Parkinson's disease (Maeda et al., 2012), and Alzheimer's disease (Cai et al., 2013). In most cases of these diseases, a direct relationship has been found between telomere shortening and a persistent high level of oxidative stress (Wolkowitz et al., 2011; Spivak et al., 2016).

Stem cells (SCs) are undifferentiated cells that can extensively proliferate (self-renew) and differentiate into many cell types in the body. The high differentiation potential of mesenchymal stem cells (MSCs) suggests that any change in their proliferation and metabolism may have unexpected consequences (Brown et al., 2019). Therefore, it seems that hMSCs can be suitable models and reference cells for evaluating the results of Faradarmani CF in biological systems.

Mohammad Ali Taheri introduced novel fields with a non-material/non-energy nature named Taheri Consciousness Fields (TCFs). From this perspective, T-Consciousness is the main component of the universe, and matter, energy, and information are derived from it. According to this theory, there are various TCFs with different functions that are non-frequency-based. These fields cannot be directly measured by science, but their effects can be investigated across various subjects through reproducible laboratory experiments. The effect of TCFs begins with the brief and immediate attention of the Faradarmangar (trained individual) to the subject of study (Taheri, 2013). This study evaluated the effect of Faradarmani CF on stem cell proliferation, telomerase activity and telomere length.

Methods and Materials

Application of the T-Consciousness Fields

TCFs were applied to the samples according to the protocols regulated by the COSMOintel Research Center (www.COSMOintel.com). More details are provided in the Common Considerations section of this issue.

Isolation and cultivation of mesenchymal stem cells

Human MSCs were isolated from a bone marrow sample of a volunteer female donor (25 years old) after obtaining informed consent. To put it briefly, the sample was washed with sterile phosphate-buffered saline (PBS) to remove debris and red blood cells. The sample was treated with 0.075% type I collagenase in PBS for 30 min at 37° C under alternating shaking. Then, collagenase I was neutralized with an equal volume of Dulbecco's Modified Eagle's Medium (DMEM) and 10% fetal bovine serum and centrifuged at 800×rpm for 10 min. The cell pellet was dissolved and cultured in DMEM high-glucose medium with 10% FBS and 1% penicillin/streptomycin. After 24 h of cultivation, non-adherent cells were discarded, and adherent cells were washed twice with PBS. After 80% confluency, the cells were passaged. After two passages, hMSCs were cultured on 6-well plates and kept in an incubator at 37 ˚C and 5% CO_2.

Cell count and telomerase tests

The morphology and number of cells were evaluated 24 hours after cultivation in Faradarmani CF-treated and control cells. After 4 days, the cells were examined under a microscope to assess contamination and cell density. When no contamination was observed, and the cells reached the appropriate confluency, they were passaged. After trypsinisation, the number of cells was counted on the hemocytometer. One mL of suspension was used to evaluate telomerase activity and telomere length.

DNA and RNA extraction

Genomic DNA extraction was performed according to the manufacturer's protocol using a kit (SinaClon, Tehran, Iran). Briefly, after the fourth and fifth passages, hMSCs were lysed in lysis buffer containing 10 mM EDTA pH 8.0, 10 mM Tris-HCl pH 8.0, 100 mM NaCl, 0.5% SDS w/v, and 100 µg/mL proteinase K for 1.5 h at 37 °C. After inactivation of proteinase K by incubation at 75 °C for 15 min, the lysate was treated with 5 µL RNase A (10 mg/mL), then centrifuged, and the supernatant was transferred to a new microtube. An equal volume of phenol:chloroform (1:1) was added and mixed thoroughly. The aqueous phase was transferred to a new tube, and genomic DNA was isolated using the ethanol precipitation method.

RNA extraction was performed using the FavorPrep™ Blood/Cultured Cell Total RNA Mini Kit (Favorgen® Biotech Corporation, Taiwan) according to the manufacturer's protocol. The concentration of RNA was measured using a NanoDrop spectrophotometer (Thermo Fisher Scientific), and samples were stored at −70 °C.

Real-Time PCR

Real-time PCR was performed to measure the relative telomere length and telomerase activity in MSCs. The master mix kit used was the Takara kit from Japan. Real-time PCR was conducted using telomerespecific primers, with 36B4 serving as the single-copy reference gene encoding acidic ribosomal phosphoprotein P0 (located on chromosome 12). The reactions were performed for 30 cycles on an Applied Biosystems 7500 system (USA).

Tel-F (5′-GGTTTTTGAGGGTGAGGGT-GAGGGTGAGGGTGAGGGT-3′), Tel-R (5′-TCCCGTATCCCTATCCCTATCCCTATC-CCTATCCCTA-3′), 36B4-F (5′-CAGCAAGT-GGGAAGGTGTAATCC-3′), and 36B4-R (5′-CCCATTCTATCATCAACGGGTACAA-3′) primers were used. The T/S ratio was calculated from the cycle threshold (Ct) values obtained for the telomere and 36B4 reactions using the comparative Ct method and was expressed as the relative telomere repeat copy number normalized to the 36B4 single-copy reference gene. In addition, telomerase activity was measured using a real-time PCR-based TRAP assay.

Data were analyzed using statistical analysis software (SPSS version 21). Data are presented as mean and standard deviation. A Student's t-test was used to compare the differences between normally distributed variables. A *p-value* < 0.05 was considered statistically significant.

Results

Results of MSCs culture

The average number of mesenchymal stem cells was counted in six wells after 24 h. The number of MSCs in the Faradarmani CF-treated group was significantly higher than that in the control group, showing an approximately 20% increase ($p < 0.05$). Morphologically, cells in the control group appeared more spread. In addition, MSCs in the treatment group appeared to detach from the plate surface more rapidly during trypsinization compared with the control group.

Results of telomere length and telomerase activity

As shown in Table 1, the effect of the Faradarmani CF on the telomere length of MSCs and the telomerase activity was statistically significant compared to the control group. Faradarmani CF significantly increased telomere length and telomerase activity in MSCs ($p < 0.05$).

Table 1. Telomere length and telomerase activity in the control and treatment group under Faradarmani CF.

Variables	Treatment group	Control group	p-value
Length of telomere	1.84±0.03	1.64±0.07	< 0.05
Telomerase activity[a]	0.10±0.005	0.09±0.002	< 0.05

Data are presented as mean ± standard deviation. [a]Telomerase activity was assessed by real-time PCR-based TRAP assay and expressed as relative activity calculated from normalized Ct values using the 2^-ΔΔCt method.

Discussion and conclusion

The aging and age-related diseases are extensively associated not only with medication but also with psychological interventions. These include proper diet, self-control, and mental and physical training to increase the telomerase activity in non-tumor cells and thus counteract age-related telomere shortening and decreased telomerase activity. These are the critical health principles that form a good way to have a long and healthy life (Epel, 2012; Farzaneh-Far et al., 2010; Spivak et al., 2016).

The present study showed that Faradarmeni CF significantly increased proliferation, telomere length, and telomerase activity in hMSCs ($p < 0.05$). Telomere length and telomerase activity are considered important biomarkers of cellular aging and potential therapeutic targets in age-related diseases, which Faradarmani CF affects, as was mentioned in this study.

As mentioned in the introduction section, TCFs, as non-material and non-energy fields, can be applied to all living and non-living beings, including plants, animals, and microorganisms, etc. Although we cannot measure TCFs directly, we can investigate their effects indirectly through various experiments.

In previous studies, we observed the effects of the TCFs on the MCF7 cancer cell line (Taheri et al., 2020a), spatial memory and avoidance behaviour of a rat model of Alzheimer's disease (Taheri et al., 2021a), wheat plant (Torabi et al., 2020), bacterial population growth (Taheri et al., 2021b), and the electrical activity of the brain during Faradarmani in the Faradarmangars population (Taheri et al., 2020b).

The present research was a study on the effect of Faradarmani CF on telomerase activity and telomere length *in vitro*, so additional research on other cell types, and *in vivo* models are required to clarify the biological relevance of these findings.

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Conflicts of Interest

The authors declare no conflict of interest.

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Ms. Laleh Amani, a distinguished, compassionate, and enthusiastic Cosmonautics researcher, passed away in 2021. We express our sincere condolences and gratitude for her extraordinary efforts in this research and pray for her peace of mind.