

Behavior of Normal and Cancerous Human Cell Lines Exposed to T-Consciousness Fields during Stress Conditions





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### **Editorial**

**Mohammad Ali Taheri**Founder of T-Consciousness Theory



### Behavior of Normal and Cancerous Human Cell Lines Exposed to T-Consciousness Fields during Stress Conditions

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Human beings have always sought to understand existence, and this pursuit has led to the resolution of many mysteries. In fact, it can be said that science, experience, and questioning form the three sides of a triangle through which human understanding and knowledge are achieved. One of the fundamental questions pertains to the concept of consciousness or awareness. How

does this phenomenon manifest? What is its function and position in humans and other living beings?

Conventional science, by studying matter and the physical aspect of the universe, has yielded valuable results. This domain can be referred to as the frequency-based world, since matter and energy, due to their wave-like nature, possess a frequency-based essence. However, in this framework, a vast portion of the cosmos lacks a frequency-based nature—such as information, consciousness, and similar phenomena. Given that conventional science naturally lacks the tools to measure and understand qualitative phenomena, a new scientific approach is needed—one that can open new windows for inquisitive minds and contribute to solving the existing mysteries.

In this regard, a new science called *Sciencefact* has been introduced to study the non-frequency-based aspects of the universe. One of the fundamental components of this domain is T-Consciousness. It is important to note that the use of scientific and laboratory tools is a common ground between Sciencefact and conventional science. In this perspective, T-Consciousness is considered a fundamental element of the universe, from which matter, energy, and information originate. Furthermore, there are various types of T-Consciousness Fields, each with different functions, all of which are subsets of the Cosmic Consciousness Network. Humans can interact with and make use of these fields. The influence of these fields begins with a brief and intentional attention (*nazar*). Interestingly, the effects of these fields can be investigated not only on living organisms but also on inanimate matter. This unique characteristic has motivated researchers across different fields to design experiments to practically test this theory.

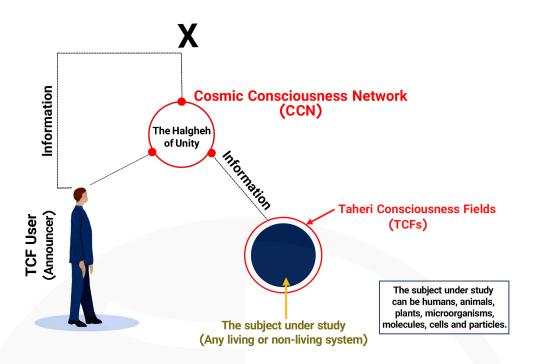
In this issue, four studies are presented. Two of these investigations explore the effects of T-Consciousness Fields on the progression of the cell cycle and cell survival under microgravity stress, using different cell lines. The results indicate that the observed changes under stress conditions differ from those seen in control conditions (normal Earth gravity). It is hypothesized that when a subject is exposed to T-Consciousness Fields, the information transmitted from these fields leads to alterations in the subject's behavior and characteristics. The differing results under normal and stress conditions not only provide empirical evidence supporting this theory but also suggest that the type of information transferred may vary depending on the environmental context. As we know, one of the

major challenges in space research is the harmful impact of reduced gravity on human health. These findings on the beneficial effects of T-Consciousness Fields offer a promising prospect for protecting the health of researchers working in this field.

The other two papers in this issue also focus on cellular studies. In one study, the viability of a breast cancer cell line was significantly reduced. In another, T-Consciousness Fields notably decreased the cytotoxicity of copper oxide nanoparticles on human blood lymphocyte cells. These observations provide further evidence of the effects of T-Consciousness Fields. Since the influence of these fields occurs without any physical intervention, the results raise an important question: how are such changes at the cellular level brought about? According to the theory of T-Consciousness, a processor is required to receive and interpret information. In other words, it is the Processing Mind that, like a manager, receives the information and subsequently alters cellular behavior. Further research using animal models, followed by clinical studies, could shed more light on the mechanisms and broader impacts of these non-physical fields.

A growing body of diverse studies is underway to investigate the behavior of living organisms and inanimate matter in response to T-Consciousness Fields. With each step forward, new dimensions of the functions and applications of these fields emerge, contributing both to the expansion of foundational concepts and to a deeper understanding of their practical implications across various scientific disciplines. It is hoped that researchers around the world will approach this subject earnestly and without prejudice, exploring it more thoroughly and helping to shape a novel and insightful perspective for the future of science.





A schematic on applying T-Consciousness Fields (TCFs). The effect of TCFs begins with connecting to the Cosmic Consciousness Network (CCN) and through the TCFs user (announcer). Variable T-Consciousness Fields are a subset of CCN, and by applying each TCF, specific information is transmitted. In this way, the subject of study, which can be living or non-living creatures, is exposed to this information. It should be noted that TCFs and the information do not have a material or energetic nature; therefore, they cannot be measured directly and quantitatively. However, it is possible to record and examine their effects by designing different experiments. For this purpose, the behavior or indicators measured by the researchers in the subject under study after being exposed to the TCFs are compared with the control samples (without the effect of TCFs), and the results are reported after statistical data analysis.

### **Considerations of This Issue**

### 1- Introduction

# 1-1 T-Consciousness and the New Science of Sciencefact

In the past few decades, the nature of Consciousness and its place in science has received considerable attention. Many philosophical and scientific theories have been presented so far in this field. In the 1980s, Mohammad Ali Taheri introduced new fields of non-material and non-energetic nature, known as T-Consciousness Fields (TCFs). In Taheri's view, T-Consciousness, along with matter and energy, are the three main constituents of the universe, with T-Consciousness being different from matter and energy. According to his theory, there are a wide variety of TCFs, with each having certain functionalities. TCFs are also considered a subset of "Cosmic Internet Network" in Taheri's theory, which is named the Cosmic Consciousness Network (CCN).

The main difference between the theory of TCFs and other concepts introduced so far for describing the nature of consciousness is the applicability and practicability of TCFs. In other words, these fields can be applied to all living organisms and non-living objects, such as plants, animals, microorganisms, materials, molecules, atoms, etc. In this respect, Mohammad Ali Taheri introduced "Sciencefact" in 2020 as one of the subgroups of the "Erfan-e-Keyhani-e-Halgheh" school, which he had previously founded. The name "Sciencefact" was chosen to confirm the existence of T-Consciousness as a "fact" scientific research method is utilized. Although mainstream science merely considers the study of matter and energy, Sciencefact investigates the effects of TCFs (which are neither material nor energy) on matter and energy and all their manifestations (such as humans, animals, plants, microorganisms, cells, materials, molecules, atoms, etc.). By repeatably conducting laboratory research experiments in various fields of science and applying TCFs, Sciencefact has emerged as a common ground between science and TCFs and uses this capability to investigate T-Consciousness and T-Consciousness Fields resulting from it.

The influence of TCFs begins with the connection (Etesal) between the Cosmic Consciousness Network as the Whole Consciousness and the subject under study as a component. The connection is established by the mind of the Faradarmangar (a person who has been trained to assign TCFs). The human mind has the role of an intermediary (announcer) that acts with short and immediate attention to the subject under study, and the main achievement is obtained due to the effects of TCFs. These fields cannot be directly measured by science, but their effects on various subjects can be investigated through repeatable experiments

### 1-2 Methodology of T-Consciousness Fields Research

The research methodology followed in the study of T-Consciousness is based on *Assumption, Argument*, and *Proof*:

The basic *Assumption* is that the universe is formed by a third element, called T-Consciousness, and that is different from matter and energy.

The *Argument* is that the existence of TCFs can be shown through their effects on matter and energy (e.g., humans, animals, plants, microorganisms, cells, materials, molecules, atoms, etc.)

The *Proof* is the scientific verification of the TCFs' effects on matter and energy (according to the Argument) through various reproducible scientific experiments

### 1-3 Study Phases in Sciencefact

To investigate and verify the existence, effects, and mechanisms of TCFs which has not been defined within mainstream science yet, the five following research phases (Phase 0 to Phase 4) and their objectives are outlined below:



In Phase 0 of the studies, the goal is to demonstrate the existence of TCFs by observing their influence on matter and energy. The nature of T-Consciousness and its definition will not be addressed in this phase. Phase 1 is dedicated to exploring various effects of different TCFs. In Phase 2, one examines the reasons behind the effects of these fields. Then, during Phase 3, the mechanisms of TCFs' effects on matter and energy are investigated. Finally, the goal of Phase 4 is to draw conclusions, particularly with regard to the *mind* and *memory of matter* and their relation to T-Consciousness, etc.

### 1-4 Using T-Consciousness Fields

The samples under study were subjected to T-Consciousness Fields (TCF) according to the specified protocol on the website of Research Management on T-Consciousness Fields (www. cosmointel.com). The request for Etesal (connection) to the cosmic consciousness network to use TCF can be submitted through the Cosmointel website in the "Assign Announcement" section. This access is freely available to everyone. Researchers can register on this website anytime to experience TCFs and conduct research in this area. Detailed information about the experiment needs to be provided to the research center; for example, the number and name of samples and controls must be specified. These studies were conducted in a double-blind manner, where lab technicians were completely unaware of TCFs theory, and the Faradarmangar at the COSMOintel research center who established the consciousness bond was unaware of the details of the study. Double-blindness is a gold standard that is common in science experiments in the field of medicine and psychology, involving theoretical and practical testing.

# Differential Effects of the Faradarmani Consciousness Field on Cell Cycle Progression of Lymphoma Raji Cells Under Clinostat-Simulated Microgravity and Earth Gravity Conditions

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### **Abstract**

This study explores a novel hypothesis. According to Taheri, various T-Consciousness Fields (TCFs) exist, each with distinct functions, and are considered subsets of the Cosmic Consciousness Network. Although these fields lack any physical entity, their effects can be detected through laboratory experiments. It is hypothesized that when a subject is exposed to TCFs, the information transmitted from these fields can alter the properties or behavior of the treated samples compared to untreated controls. In the present experiment, the Faradarmani Consciousness Field, one type of TCF, was applied to investigate its effect on the cell cycle progression of the lymphoma Raji cell line under both simulated microgravity and normal Earth gravity conditions. There were four experimental groups, and the experiment duration was 48 hours. Samples not exposed to the Faradarmani Consciousness Field were considered the control group. Based on flow cytometry analysis, apoptosis was observed in cells exposed to microgravity (MG), with the sub-G1 phase increasing to approximately 42% (p-value< 0.05), whereas Faradarmani-treated samples remained almost unchanged under MG stress Similarly, Faradarmani-treated samples exhibited similar percentages of G1 and S phases under microgravity conditions compared to Earth gravity, while a significant decrease was observed in samples without the field effect (p-value < 0.05). However, under normal gravity conditions, the effect of FCF was not significant compared to the control. These observations suggest that the Faradarmani Consciousness Field influences cell cycle progression differently depending on environmental conditions. Under microgravity (MG) stress, the information transmitted from this T-Consciousness Field appears to have an alleviating effect, whereas under normal Earth gravity, it did not produce a significant change.

Keywords: Faradarmani, Microgravity, Information, Cell Cycle, Raji, Apoptosis, Clinostat



### Introduction

Human curiosity has always been a driving force behind exploration and discovery. From ancient civilizations observing the stars to modern scientists launching satellites and rovers, the desire to understand the universe has been deeply rooted in our nature. Despite the wonders of space, exploration has always come with immense physical risks for astronauts. For example, microgravity stress leads to muscle atrophy, bone density loss, cardiovascular changes, and altered immune responses (Baran et al., 2002; Manna et al., 2024). Although countermeasures like treadmill and cycling exercises, along with special suits to stress skeletal muscles, are used to prevent physical deterioration in microgravity, they are not fully effective. Despite these efforts, the negative effects of weightlessness often persist after returning to Earth, leading to reduced work efficiency and quality of life (Ruden et al., 2018; Scott et al., 2019; Bonanni et al., 2023).

Using terrestrial tools like the clinostat is crucial for advancing our understanding of how microgravity affects biological systems, without the high cost and complexity of space missions (Ferranti et al., 2021). A clinostat is a device that slowly rotates biological samples, such as cells or small organisms, to cancel out the directional influence of gravity over time (Kim et al., 2023). This creates a condition that mimics the effects of microgravity. It enables researchers to study how living systems respond to the absence of gravitational force, similar to what occurs during spaceflight (Kiss et al., 2019).

T-Consciousness is a term coined by Mohammad Ali Taheri, proposing that consciousness is not an emergent property of matter; rather, information, matter, and energy originate from it. Within framework. various T-Consciousness this Fields (TCFs)—each with distinct functions are introduced as subsets of the Cosmic Consciousness Network (Taheri, 2013). Unlike mainstream theories such as Integrated Information Theory (IIT), Global Workspace Theory (GWT), Higher-Order Thought (HOT) Theory, or Orch-OR, which attribute the source of consciousness to physical structures (e.g., the brain, information integration, or quantum processes) (Sattin et al., 2021), TCF theory suggests that the brain functions primarily as a detector of information—hardware operating in conjunction with the software aspect, the mind. Indeed, instead of focusing on neural complexity in explaining conscious experience, TCF theory adopts a non-local perspective, viewing T-Consciousness as independent of neural activity and beyond the brain.

Moreover, what makes this theory unique is its practical and testable model of non-physical field influence. Various experimental studies have reported the effects of TCFs on a wide range of subjects, including plants, animals, cells, and microorganisms (Taheri et al., 2022; Torabi et al., 2023; Taheri et al., 2024). It is hypothesized that information transmitted from TCFs can modify the properties and behavior of the studied subjects. In the current experiment, the effects of the Faradarmani Consciousness Field (FCF) on cell cycle progression of the lymphoma Raji cell line were evaluated to investigate whether this treatment could mitigate the adverse effects of microgravity stress induced by a clinostat device.

### Material and methods

# Faradarmani Consciousness Field application

FCF was applied to the samples according to protocols regulated by the COSMOintel research center (www.COSMOintel.com). Further details are provided in the general considerations of this issue.

### Microgravity (MG) application

In this study, the microgravity condition was simulated using a clinostat, which was donated by the United Nations Office for Outer Space Affairs in Vienna to the Aerospace Research Institute of Iran. The clinostat was sterilized using ultraviolet light and 70% ethanol, and

subsequently placed in an incubator set at 37 °C (Figure 1). The experiment included four groups (n = 3) as follows: Group 1 – microgravity with FCF (MG + FCF); Group 2 (control) –

microgravity without FCF (MG); Group 3 – normal gravity with FCF (1G + FCF); and Group 4 (control) – normal gravity without FCF (1G).





Figure 1. The clinostat used in this experiment and the placement of samples under microgravity (MG) and normal gravity (1G) conditions.

### Cell culture

In this experiment, human B-lymphoblastoid Raji cells were purchased from the Pasteur Institute of Iran and cultured in Roswell Park Memorial Institute 1640 medium containing 10% fetal bovine serum (Gibco Laboratories, Grand Island, NY), 100 IU/ml penicillin, and 100 µg/ml streptomycin. The cultures were maintained in a humidified incubator at 37 °C (Memmert, Schwabach, Germany) with 5% CO<sub>2</sub>. To prevent the formation of air bubbles, the flasks were completely filled with culture medium, and then the samples were secured in a circular holder within the clinostat environment. The rotational speed was set at 30 rpm and continued for 48 hours to cover the cell doubling time.

### Flow cytometry

Harvested cells of these two experiments were washed twice with PBS, and after adding 50 μl of cold PBS (+2 to +8 °C), they were suspended with a short vortex period. Then cells were fixed in 1ml cold 70% ethanol (-20 °C), and

were resuspended using the vortex mixer. After that, the cell suspension was centrifuged at 1500 rpm for 20 minutes, at room temperature, and after removing the supernatant; cells were washed once with PBS. Next, the PBS was slowly removed and 1ml MIX MASTER PI solution was added. The final concentration of cells in mentioned solution must be  $5 \times 10^5$ cells/ml. Finally, the cells are incubated for 30 minutes at ambient temperature and read by flow cytometry. The proportion of cells at different stages of the cell cycle was assessed using a flow cytometer in the BD FACS Calibur system (BD bioscience, San Joe, CA, USA). The cell cycle phases were assessed by FlowJo software (Tree Star, San Carlos, CA).

Master PI mix solution for cell cycle

Propidiumlodide (PI) 1 mg/mL: 40 µl

RNase (DNaseFREE) 10mg/mL: 10 µl

PBS, ca<sup>+2</sup>, mg<sup>+2</sup> Free: 950 µl



### Data analysis

Each experiment was repeated three times, and the data were presented as mean ± standard error (SE). One-way analysis of variance (ANOVA) followed by multiple comparisons with a 95% confidence interval was performed using GraphPad Prism software (version 9), and p-values less than 0.05 were considered statistically significant.

### **Results and Discussion**

Table 1 shows the percentage distribution of cell cycle phases with and without Faradarmani Consciousness Field (FCF) treatment under both microgravity (MG) and Earth gravity (1G) conditions. As shown, the lowest average percentage of live cells within the selected gate was observed under microgravity (MG) stress without FCF treatment. Similarly, analysis of cell cycle progression reveals a significant increase in the sub-G1 phase in this group. In contrast,

the FCF-treated sample under MG conditions displays a sub-G1 percentage comparable to that observed under Earth gravity (1G).

A flow cytometry plot displaying FL3-A versus SSCH parameters enables the analysis of cellular populations based on DNA content and internal complexity (McKinnon, 2018). Figure 2 shows that elevated SSCH values in the untreated microgravity (MG) sample suggest the induction of structural damage or apoptosis.

Figure 3 illustrates the cell cycle phase distribution based on DNA content. The black curve in the figure (a) displays a distinct peak at DNA content values <200, which is characteristic of the sub-G1 phase, typically associated with apoptotic cells containing fragmented DNA (Plesca et al., 2008).

Table 1. Effect of Faradarmani Consciousness Field (FCF) on the percentage of cell cycle phases under microgravity (MG) or earth gravity (1G) condition.

Groups	Samples description	The average percentage of live cells in selected gate	Sub G1	G1	S	G2	Super G2
FCF-/MG	Control - MG condition without FCF	64	42.10±2.44	20.33±0.80	28.27±0.92	11.17±1.88	0.70±0.31
FCF+/MG	MG condition with FCF	78	6.29±0.79	47.78±0.05	40.85±1.96	10.17±1.13	0.57±0.33
FCF-/1G	Control – 1G condition without FCF	77	6.90±2.90	47.78±6.37	39.12±1.61	9.10±1.44	0.41±0.02
FCF+/1G	1G condition with FCF	78	6.79±1.41	42.66±1.79	42.21±2.43	11.62±1.61	0.59±0.22

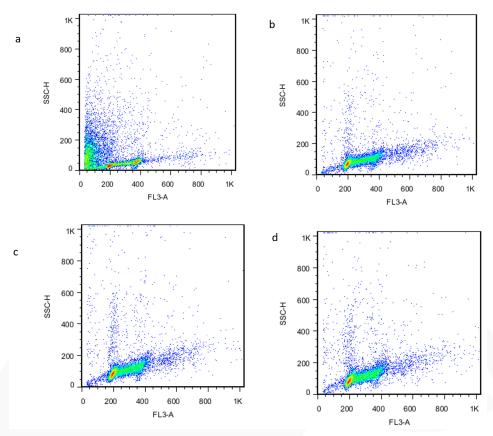


Figure 2. Representative FL3-A histograms of Raji cells under (a) Microgravity (MG) without Faradarmani Consciousness Field(FCF), (b) MG with FCF, (c) 1G without FCF, and (d) 1G with FCF.

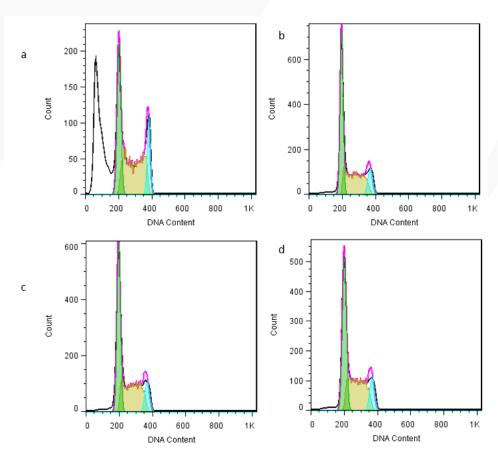


Figure 3. Representative histogram (single replicate) showing cell cycle phase distribution in Raji cells under (a) MG without FCF, (b) MG with FCF, (c) 1G without FCF, and (d) 1G with FCF.



Furthermore, statistically significant changes in cell cycle phases are presented in Figure 4. As shown, MG stress led to a marked increase in the sub-G1 population (p-value< 0.05) and a significant reduction in G1 and S phases

in untreated samples (p-value< 0.05). In contrast, FCF-treated samples displayed values comparable to the controls under Earth gravity (1G) conditions.

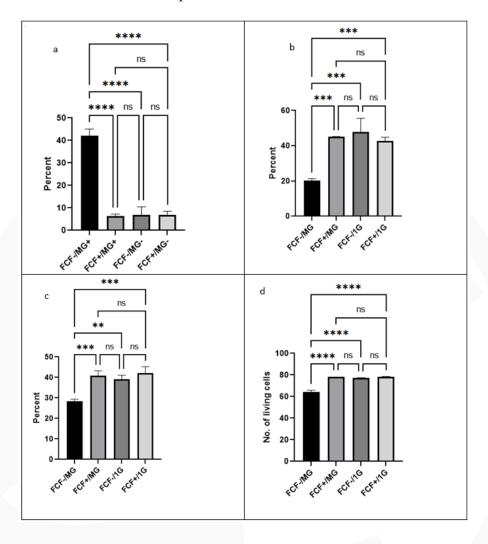


Figure 4. Effect of Faradarmani Consciousness Field (FCF) and gravity on the percentage of the living cells in (a) Sub-G1, (b) G1, and (c) S phases, as well as the number of live cells (d). MG: microgravity; 1G: Earth gravity; ns: not significant; \*\*p < 0.001; \*\*\*p < 0.0001; \*\*\*\*p < 0.00001.

Numerous studies on various cell types have demonstrated morphological sensitivity to microgravity (Tauber et al., 2017; Bradbury et al., 2020). For example, one study reported that simulated microgravity significantly disrupts the mechanical properties and cytoskeletal structure of endothelial cells. After just 24 hours of exposure, these cells showed a marked reduction in stiffness and viscosity (Janmaleki et al., 2016). This stress has also been reported to impair cell cycle progression (Vidyasekar

et al., 2015) and increase apoptosis (Pan et al., 2020; Sokolovskaya et al., 2020).

It is well established that physical forces such as gravity have played a fundamental role in the evolution of life on Earth, influencing numerous biological processes (Topal and Zamur, 2021). In this study, cells treated with the FCF tolerated reduced gravity and remained viable. As noted in the introduction, it is hypothesized that FCF transmits information capable of altering the properties or behavior of the treated system.

The current results demonstrate that FCF substantially mitigated the damaging effects of microgravity (MG), while FCF-treated samples under Earth gravity showed responses similar to untreated controls.

This suggests that the information conveyed by FCF may vary depending on environmental conditions. Consistent with its alleviative effects, these findings align with our previous studies showing that FCF reduced the harmful impact of salinity on wheat seedlings and decontaminated radioactive materials in aquatic environments (Taheri et al., 2022b; Torabi et al., 2023). This observation warrants further investigation under real weightlessness, as well as hypergravity conditions, to gain deeper insight into how FCF treatment may influence the behavior of organisms in space.

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# T-Consciousness Fields Inhibit Apoptosis and Promote ATP Production in HEK-293 Cells Under Microgravity Stress

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### **Abstract**

7-Consciousness Fields (TCFs) are introduced as non-physical entities, with their effects initiated ▲ through the human mind. It is hypothesized that information transmitted via TCFs may influence the behavior of studied subjects. The present experiment aimed to investigate the effects of TCFs on cell cycle progression and ATP production in the HEK-293 cell line under both microgravity (MG) and Earth gravity (1G) conditions. To achieve this, cultured cells were exposed to TCFs for 24 hours following an initial 24-hour incubation period, under either microgravity (MG) conditions, simulated by clinostat rotation, or a standard Earth gravity (1G) environment. Untreated cells served as the control group. Flow cytometry was used to assess the distribution of cells across different phases of the cell cycle, and ATP concentration was measured by evaluating luciferase enzyme activity. The results showed a significant increase in the sub-G1 phase under microgravity (MG) conditions (p < 0.05), indicating elevated apoptosis. In contrast, TCFs-treated samples maintained sub-G1 levels comparable to those under Earth gravity (1G). Additionally, TCFs treatment significantly increased the S phase under MG (p < 0.05) and the G2 phase under 1G (p < 0.05). ATP concentrations were markedly reduced under MG in both treated and untreated samples compared to 1G (p < 0.05). However, in the clinostat environment, ATP levels in TCFs-treated samples were approximately twice as high as those in untreated controls (p < 0.05), whereas no significant difference was observed under 1G conditions. These findings suggest that the reduced sub-G1 phase in TCFs-treated samples under MG reflects apoptosis inhibition, while increased ATP levels and prolonged S phase indicate enhanced cellular efficiency under stress. The distinct outcomes under MG and 1G imply that the information transmitted by TCFs may vary depending on environmental conditions. Further studies are needed to elucidate the underlying mechanisms of TCF action.

**Keywords:** T-Consciousness Field, Microgravity, Clinostat, Cell cycle, Information

### Introduction

It is well-documented that reduced gravity can have adverse effects on human physiology (Mochi et al., 2022). Indeed, space exploration has presented scientists with numerous challenges, many of which stem from microgravity-induced alterations in cellular responses. Consequently, researchers have long sought therapeutic interventions to safeguard the health of astronauts during extended space missions (Nguyen et al., 2021). One promising approach to understanding the impact of microgravity on biological systems is the investigation of cellular parameters such as proliferation, apoptosis, and cell cycle progression (Sokolovskaya et al., 2014).

The cell cycle, a fundamental process in eukaryotic cells, has been the focus of over a century of research. It consists of four main phases: G1, S, G2, and M (Wang, 2022). Disruptions in cell cycle regulation, as well as in programmed cell death mechanisms, can lead to irreversible health consequences and are implicated in the development of various disorders, including cancer, inflammation, neurodegenerative diseases, and cardiovascular conditions (Wiman and Zhivotovsky, 2017).

Life on Earth has evolved under constant gravitational force, and the human body is naturally adapted to this terrestrial environment (Adamopoulos et al., 2021). As a result, space travel introduces a range of physiological threats. For instance, alterations in the extracellular microenvironment of cancer cells under microgravity conditions can stimulate the secretion of cytokines and tumor growth factors, thereby potentially increasing cancer malignancy (Kim et al., 2021). Moreover, significant changes in the expression of genes related to oxidative stress, inflammation, and cell cycle regulation have been observed in mice during the STS-131 space shuttle mission, which may contribute to cardiac dysfunction (Kumar et al., 2021).

According to Taheri, T-Consciousness is a fundamental element of the universe. Within this framework, various T-Consciousness Fields (TCFs) exist, each serving distinct functions as subcategories of the Cosmic Consciousness Network. These fields cannot be measured using conventional quantitative instruments; however, their effects can be observed and recorded through laboratory experiments conducted on a range of subjects. The influence of TCFs is initiated through the human mind, requiring only a brief moment of attention, typically just a few seconds (Taheri, 2013).

These fields can be applied to both living organisms and inanimate materials. In our previous studies involving animal, plant, and cell models, we observed that the properties and behaviors of TCF-treated samples were noticeably altered compared to untreated controls. It is hypothesized that these observed changes result from the transmission of information from the TCFs (Taheri et al., 2023; Torabi et al., 2023; Taheri et al., 2024). The aim of this study was to investigate the effects of three types of T-Consciousness Fields (TCFs) on cell cycle progression and ATP production in the HEK-293 cell line under both microgravity (MG) and normal Earth gravity (1G) conditions.

### Material and methods

### **T-Consciousness Fields application**

Three types of TCFs, including TCF1, 2 and 3, were applied to the samples according to protocols regulated by the COSMOintel research center (www.COSMOintel.com). Further details are provided in general consideration section of this issue.

### Microgravity (MG) application

In this study, microgravity (MG) conditions were simulated using a clinostat, which was donated by the United Nations Office for Outer Space Affairs in Vienna to the Aerospace Research Institute of Iran. Although the gravity vector cannot be eliminated on Earth's surface.



MG (approximately 10<sup>-6</sup> g) can be effectively simulated using this device. The clinostat was first sterilized with ultraviolet light and 70% ethanol, then placed inside an incubator set to 37 °C. After an initial 24-hour incubation period, cultured cells were treated with T-Consciousness Fields (TCFs) under either MG or normal Earth gravity (1G) conditions. Four experimental groups were established (n = 3 per group): MG with TCFs (Group 1), MG without TCFs as control (Group 2), 1G with TCFs (Group 3), and 1G without TCFs (Group 4). The samples without TCFs were considered as control. The TCFs treatment was initiated simultaneously with clinostat rotation and continued for 24 hours

### **Cell Culture**

In this study, the HEK-293 cell line—characterized by epithelial morphology and originally derived from the kidney of a human embryo—was obtained from the Pasteur Institute of Iran. Cells were cultured in a 6-well plate (4 × 10<sup>5</sup> cells per well) using Dulbecco's Modified Eagle Medium (DMEM) supplemented with 10% fetal bovine serum (FBS) and 1% penicillin/streptomycin. Incubation was performed at 37 °C in a humidified atmosphere containing 5% CO<sub>2</sub>.

After 24 hour, the medium in each well was removed, and the wells were washed with phosphate-buffered saline (PBS). A 1X solution of 0.25% trypsin-EDTA was then added to each well. The plate was incubated at 37 °C for 5 minutes to allow cell detachment. Trypsin activity was neutralized by adding 300 µl of complete culture medium. The cells were then collected, centrifuged at 1200 rpm for 5 minutes, and the resulting cell pellet was stored at -80 °C until ATP assay analysis. ATP concentration was measured using a luminometer (Berthold Technologies GmbH & Co. KG) based on luciferase enzyme activity.

In this study the following materials were used: ATP (Roche), D-luciferin potassium salt (Resem, The Netherlands), Fetal bovine serum

(FBS) (BIO-IDEA), Dulbecco's modified Eagle's medium (DMEM) (BIO-IDEA), Penicillin/streptomycin (BIO-IDEA), Trypsin-EDTA 25% (BIO-IDEA), Tris-HCl (Merck), NaOH (Merck), MgSO4 (Merck), PMSF

### **Cell Lysis**

To lyse the cells, 30 µl of CCLR buffer (containing 50 mM Tris, 150 mM NaCl, 1% Triton X-100, and 0.1 mM PMSF; pH 6.9) was added to the cell pellets. The samples were incubated on ice for approximately 20 minutes, followed by centrifugation at 13,000 rpm for 15 minutes at 4 °C. The resulting supernatant was then collected and used for the ATP assay.

### ATP assay

An ATP standard curve was first established by preparing a serial dilution of ATP within the concentration range of 0.001–1 mM. To eliminate any ATP contamination, the luciferase enzyme was dialyzed in 50 mM Tris buffer for 24 hours. For ATP quantification in the treated cells, equal volumes (1:1:1 ratio) of the dialyzed luciferase enzyme, luciferin, and cell lysate were mixed in a tube. The resulting luminescence was measured using a luminometer (Jouaville et al., 1999).

### **Flow Cytometry**

Harvested cells were washed twice with PBS and resuspended in 50 µl of cold PBS (2-8 °C) using brief vortexing. The cells were then fixed in 1 ml of cold 70% ethanol (-20 °C) and thoroughly mixed with a vortex mixer. Following fixation, the cell suspension was centrifuged at 1500 rpm for 20 minutes at room temperature. After discarding the supernatant, the cells were washed once with PBS. Subsequently, PBS was carefully removed, and 1 ml of MIX MASTER PI solution was added, ensuring a final cell concentration of  $5 \times 10^5$  cells/ml. The cells were then incubated at room temperature for 30 minutes before analysis. The distribution of cells across different stages of the cell cycle was assessed using a BD FACSCalibur flow

cytometer (BD Biosciences, San Jose, CA, USA). Data analysis was performed with FlowJo software (Tree Star, San Carlos, CA, USA).

Master PI mix solution for cell cycle

Propidium iodide (PI) 1 mg/ml: 40 μl; RNase (DNaseFREE) 10mg/mL: 10 μl; PBS, ca+2, mg+2 Free: 950 μl.

### **Statistical Analysis**

Each experiment was conducted in triplicate, and the data are presented as mean  $\pm$  standard error (SE). Statistical analysis was performed using one-way analysis of variance (ANOVA), followed by multiple comparisons with a 95% confidence interval, using GraphPad Prism software (version 8). Differences were considered statistically significant at p < 0.05.

### **Results and Discussion**

As shown in Table 1, the highest percentage of live cells based on the FSC channel was observed in the TCF-treated samples under microgravity (MG) conditions, indicating a resistance to MG-induced stress. Additionally, Figure 1 (a) shows

that elevated SSC-H values in the untreated microgravity (MG) sample may indicate the induction of structural damage or apoptosis. Assessing changes in the Sub-G1 phase of the cell cycle provides insight into whether cells are healthy or undergoing apoptosis. In the clinostat environment, the proportion of cells in the Sub-G1 phase was approximately twice that of the control under 1G conditions.

Environmental stress factors in space such as MG can pose a threat to the DNA integrity of living organisms (Moreno-Villanueva et al., 2017) and induce apoptotic cell death (Singh et al., 2021; Kossmehl et al., 2003). Apoptotic cells are usually associated with morphological changes and reduced DNA content, which can be detected by flow cytometry (sub-G1 DNA content) (Plesca et al., 2008). In such conditions, DNA repair pathways are activated, including cell cycle arrest, which provides time for DNA repair (Zhou and Elledge, 2000; Prasad et al., 2020). In the current study, an increase in the sub-Glwas not observed in the TCF-treated samples. These findings suggest that TCF treatment may mitigate the adverse effects of MG stress on cell cycle progression.

Table 1. Effects of T-Consciousness Fields (TCFs) on the distribution of cell cycle phases under microgravity (MG) and Earth gravity (1G) conditions.

Sample	Live cells based on FSC channel (%)	Sub G1	G1	S	G2	Super G2
MG-Control	61.85±2.83	13.41±0.86	41.14±1.15	28.44±2.47	23.17±0.02	0.61±0.45
MG-TCFs	74.70±2.87	7.49±0.21	38.61±0.60	33.70±1.65	21.89±0.27	$0.42{\pm}0.08$
1G-Control	67.70±1.56	8.96±1.94	43.28±3.71	29.94±0.45	19.89±2.63	$0.80 \pm 0.04$
1G-TCFs	67.40±2.25	$8.68\pm0.40$	39.19±1.36	$29.43 \pm 0.05$	25.63±0.76	$0.56 \pm 0.34$



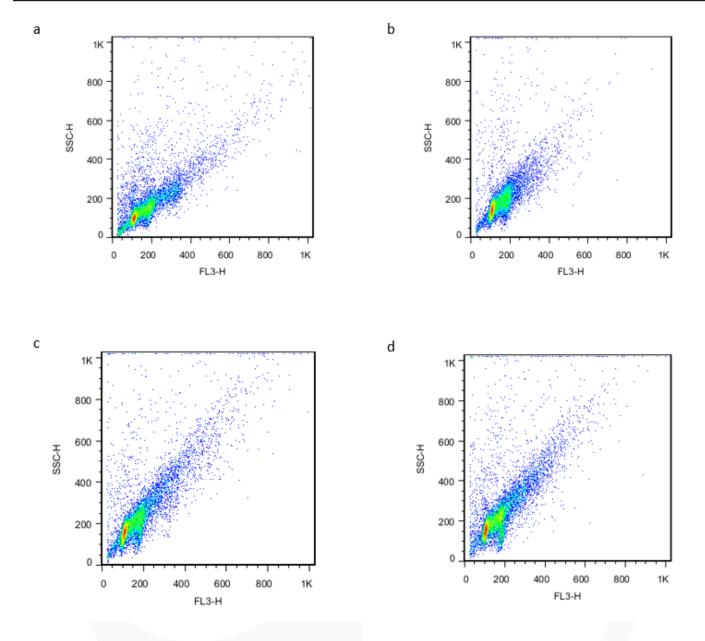


Figure 1. Representative histograms showing flow cytometry analysis using the FL3-A channel. (a) Microgravity (MG) without TCFs treatment, (b) MG with TCFs treatment, (c) 1G without TCFs treatment, (d) 1G with TCFs treatment.

Statistically significant changes are shown in Figure 2. TCFs treatment resulted in a downward trend in the G1 phase and a significant increase in the S phase. Previous studies have shown that MG stress can induce G1 phase arrest, leading to impaired cell cycle progression and reduced cellular proliferation (Quynh Chi et al., 2020; Hoang et al., 2025). In the present study, the observed increase in the S phase, accompanied by a decline in the G1 phase, suggests that TCFs treatment facilitates the transition from G1 to S phase under MG conditions. This indicates a potential protective or adaptive role of TCFs in

promoting cell cycle progression and mitigating MG-induced cell cycle arrest.

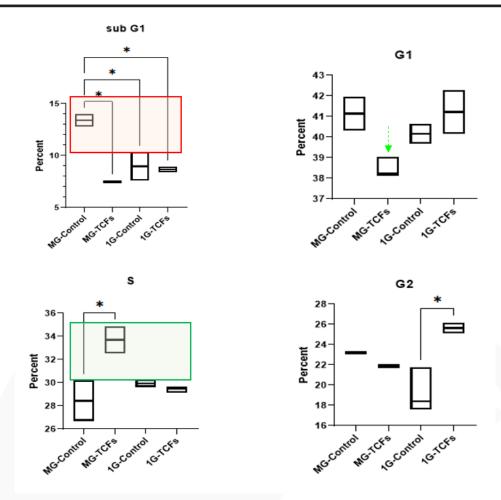


Figure 2. Box plot showing the effect of T-Consciousness Fields (TCFs) on the percentage distribution of cell cycle phases. Green and red boxes indicate changes in trend relative to the 1G control. \*: p-value < 0.05. MG: microgravity

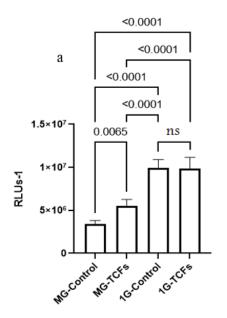
The behavior of cells under the influence of TCFs in normal Earth gravity differed from that observed under microgravity conditions. Notably, the longest G2 phase was found in TCFstreated cells under 1G. The G2 phase is the final part of interphase, during which cells prepare for mitotic division by duplicating organelles, such as mitochondria, and synthesizing proteins essential for cell division (Mascanzoni et al., 2019). Given that no significant changes were observed in the S phase or in overall cell proliferation in TCFs-treated cells under 1G, the prolonged G2 phase may reflect a form of cell cycle arrest. This finding warrants further investigation into organelle dynamics and other cellular changes under TCFs treatment to better understand the underlying mechanisms.

The concentrations of ATP and luciferase activity in the experimental groups are presented in Table 2 and Figure 3. MG stress significantly reduced ATP production in both the control and TCFs-treated samples, with an approximate 96% decrease in the control group compared to the 1G control. However, under MG conditions, TCFs treatment markedly increased ATP levels—by up to 115%—compared to the untreated MG control. In contrast, TCFs treatment under Earth gravity (1G) did not lead to a significant change in ATP concentration.



Table 2. Changes in luciferase activity and ATP concentration with and without T-Consciousness Fields (TCFs), compared to their levels under Earth gravity (1G).

Sample	Control/1G	TCFs+/1G	Control/MG	TCFs+/MG
Luciferase activity Mean/ RLU.s <sup>-1</sup>	9,966,667	9,833,333	3,383,333	5,540,000
Std. Deviation	1,322,876	927,362	440,076	733,485
[ATP]Mean /μM	9.472	9.194	0.379	0.816
Std. Deviation	1.608	2.009	0.049	0.185
([ATP] Control 1G) ([ATP] Sample)	1	1.03	24.99	11.61



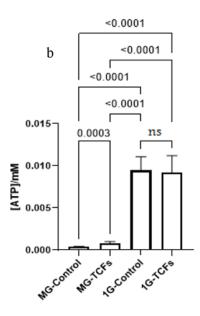


Figure 3. Effects of T-Consciousness Fields (TCFs) on a) the activity of luciferase enzyme and b) the concentration of ATP under microgravity (MG) and Earth gravity (1G) conditions. Vertical bars indicate mean  $\pm$  standard error of three replicates. P-value < 0.05 considered as significant, and ns shows non-significant change.

Microgravity stress markedly decreased ATP concentrations compared to the 1G condition. As shown in Table 2, ATP levels under MG were approximately 25-fold and 12-fold lower in the control and TCF-treated cells, respectively. Despite this significant reduction, the percentage of viable cells was not significantly affected. Notably, although ATP concentration in TCF-treated cells under MG decreased by about 91% relative to the 1G control, cell viability was approximately 6% higher than under normal gravity. We hypothesize that, in the absence of

gravity, cells may exhibit improved metabolic efficiency or altered energy utilization mechanisms.

This observation is consistent with several studies suggesting that, in the absence of gravity, cells may adapt by optimizing energy utilization and potentially requiring less ATP to maintain essential functions. For example, plants grown under microgravity conditions have shown altered gene expression related to cell wall synthesis, oxidative stress responses, and energy

metabolism—adaptations that may help sustain growth and development (Soleimani et al., 2019; Baba et al., 2022). Similarly, certain bacterial species exhibit gene expression changes that enhance their resilience to microgravity stress, potentially contributing to more efficient energy utilization (Milojevic et al., 2020).

### **Conclusion**

In conclusion, this study showed that TCFs differentially affect cell cycle progression and ATP production in HEK-293 cells under simulated microgravity (via clinostat) and Earth gravity conditions. While microgravity stress induced an increase in the sub-G1 phase—

indicating apoptosis—TCFs-treated exhibited a pattern more similar to those in normal gravity. Furthermore, TCFs treatment led to significantly higher ATP levels under microgravity, suggesting enhanced energy utilization or protection against MG-induced metabolic disruption. What if TCFs can prevent the damaging effects of microgravity on human cells? TCFs may offer a novel, non-invasive strategy to support astronaut health in space. Further studies in real microgravity and even hypergravity environments are needed to explore this potential and determine whether TCFs could become a valuable tool in future space exploration.

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# The Impact of Faradarmani Consciousness Field on Controlling the Toxicity of Copper Nanoparticles on Lysosomal Membrane Rupture in Human Blood Lymphocytes

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### **Abstract**

This study examines the effect of one type of 1-consciousness Field (FCF), on mitigating the toxicity of copper oxide nanoparticles (CuO-This study examines the effect of one type of T-Consciousness Field, named Faradarmani NPs) in human blood lymphocytes. Nanoparticle toxicity, particularly its adverse effects on cellular systems, is a critical concern in nanotechnology. FCF, introduced by Mohammad Ali Taheri, is described as a novel field, neither matter nor energy, and is non-quantifiable. However, its effects can be assessed through standard scientific experiments. In this research, the toxicological effects of CuO-NPs with and without FCF treatment were analysed by evaluating lysosomal membrane integrity and cell viability. Lymphocytes were isolated from healthy individuals' blood and exposed to CuO-NPs at various concentrations for six hours. Results demonstrated that CuO-NPs reduced cell viability in a dose-dependent manner, with an IC50 of 420 µM. The toxicity was linked to oxidative stress, leading to lysosomal membrane rupture and decreased cell survival. Under the influence of FCF, lymphocytes showed improved survival rates despite CuO-NP exposure. FCF significantly (p-value< 0.05) reduced lysosomal membrane damage and increased cell viability. Interestingly, higher nanoparticle concentrations in the presence of FCF corresponded to enhanced cell survival compared to untreated controls. This study highlights the potential protective effect of FCF against the toxicity of nanoparticles, paving the way for further research into their mechanisms and applications in biomedical science.

Keywords: Faradarmani Consciousness Field, CuO nanoparticles, Lysosomes, Toxicology

### Introduction

Nanotechnology has revolutionized numerous fields, from medicine to environmental science, by offering innovative solutions at the nanoscale. Among these advancements, copper oxide nanoparticles (CuO-NPs) have garnered significant attention due to their unique properties and wide applications. The pharmaceutical industry has increasingly embraced nanotechnology to enhance drug delivery (especially for cancer therapy), and therapeutic efficacy (Gaetke and Chow, 2003).

Copper oxide nanoparticles (CuO-NPs) have gained significant interest due to their diverse applications across various fields. In pharmaceuticals, they enhance drug solubility, stability, and controlled release, and their ability to generate reactive oxygen species (ROS) has been utilized in cancer treatments for selectively targeting tumor cells (Assadian et al., 2018). They are also being explored as contrast agents in medical imaging techniques such as MRI and fluorescence imaging. Additionally, CuO-NPs are used in biosensors for detecting biomolecules and pathogens, as well as in batteries, fuel cells, and photocatalysis due to their excellent electrical and thermal conductivity. In environmental applications, they aid in wastewater treatment by degrading organic pollutants through photocatalysis, while in agriculture, they serve as antimicrobial agents in pesticides and fertilizers, protecting crops from infections and facilitating the controlled release of nutrients to improve soil fertility (Patra et al., 2018).

Despite their advantages, the use of CuO-NPs in medicine raises concerns about toxicity, as they can induce oxidative stress, damage cellular structures, and cause lysosomal membrane rupture in human blood lymphocytes, leading to cell damage and reduced viability (Ahmed et al., 2010). To address these challenges, recent research has focused on mitigating nanoparticle-induced toxicity through approaches such as coating CuO-NPs with biocompatible materials to reduce surface reactivity while maintaining their functionality (Hussain et al., 2016).

Studies on exposure to CuO-NPs have shown that it leads to oxidative stress, DNA damage, growth inhibition in organisms, and cell death (Assadian et al., 2019). The toxicity of various nanomaterials has been investigated in mice. It was demonstrated that CuO-NPs, compared to titanium (Ti), iron (Fe), or silver (Ag) oxides, cause stronger inflammation in addition to increased total cell and neutrophil uptake into the lungs, elevated total protein levels, and lactate dehydrogenase activity in Broncho alveolar lavage (BAL) fluid. Moreover, along with evidence of inflammation induced in mice exposed sub-acutely to CuO-NPs, these nanoparticles are considered one of the key toxic nanomaterial's for mammals (Grassian et al., 2007).

Few studies have examined the adverse effects of CuO on the immune system, and the potential toxic impact of CuO-NPs on the human immune system has yet to be thoroughly investigated. This research aimed to study the effect of CuO-NPs on isolated human lymphocytes, as adverse effects on these crucial components of the immune system could disrupt its overall functioning, and investigated the possible role of Faradarmani Consciousness Field (FCF) in controlling CuO-NP toxicity by assessing their impact on lysosomal membrane integrity and lymphocyte survival. To determine the underlying effect of CuO-NP cytotoxicity, we studied the effects of half IC50, IC50, and 2IC50 concentrations of CuO-NPs on various cellular and intracellular factors. The IC50 of CuO nanoparticles refers to the concentration that kills 50% of lymphocytes during a 12hour treatment. In the sample, all conditions are identical to the control, with the only difference being that all processes are influenced by FCF.

Given their extensive applications, it is crucial to explore strategies for minimizing nanoparticle-induced cellular damage to ensure safer use in advanced medicines. Since CuO-NPs can enter the body through the skin, eyes, or inhalation, their toxic effects are unavoidable (Fahmy and Cormier, 2009). Therefore, developing methods to control their toxicity and mitigate



their harmful effects on humans and biological systems, including animals, is essential (Siddiqui et al., 2013).

### **Materials and Methods**

# Faradarmani Consciousness Field Application

Faradarmaniwasappliedtothesamplesaccording to protocols regulated by the COSMOintel research center (www.COSMOintel.com). More details are presented in general consideration of this issue. The samples without Faradarmani Consciousness Field application served as controls and for treated samples, this field was announced once at the beginning of the experiment.

### Chemicals

Acridine orange, CuO nanoparticle, and trichloroacetic acid were purchased from Sigma-Aldrich Co. RPMI1640 and FBS (fetal bovine serum) were purchased from Gibco, Life Technologies, Grand Island, NY., bovine serum albumin, Ficoll-Paque PLUS was obtained from GE Healthcare Bio-Science Company.

# Isolation and Treatment of Human Lymphocytes

Lymphocytes were obtained from the blood of 20 healthy donors aged 18 to 40 years who showed no signs of infectious disease at the time of sample collection. After dilution with an equal volume of phosphate-buffered saline (PBS), the blood was layered over 3 mL of Ficoll-Paque and centrifuged at 2000 × g for 20 minutes. The plasma layer was carefully removed, and the lymphocyte-containing layer (the buffy coat) was transferred to a new 15 mL tube. The transferred cells were diluted with 10 mL PBS and centrifuged at 1000 × g. The supernatant was discarded.

After the washing step, lymphocytes were suspended in red blood cell lysis buffer (150 mM NH4Cl, 10 mM NaHCO3, 1 mM EDTA,

pH 7.4) and incubated at 37°C for 5 minutes. The cells were washed twice with PBS, and approximately 1–1.5 × 10<sup>6</sup> cells were resuspended in 1 mL of RPMI-1640 medium containing 10% FBS and 1% antibiotic (penstrep) for further experiments. To assess cell viability, lymphocytes were treated with 0.1–5 mM CuO nanoparticles (CuO-NPs) for 12 hours. Additional experiments were performed by incubating lymphocytes with 0.2, 0.4, and 0.8 mM CuO-NPs for 2, 4, and 6 hours.

### **Cell Viability Assay**

The trypan blue exclusion dye was used to evaluate cell viability, as it can penetrate the cell membrane and label dead cells. Approximately 1 × 10<sup>4</sup> human lymphocytes per well were cultured in 96-well plates and treated with different concentrations of CuO-NPs. An equal volume of the cell suspension was mixed with 0.4% trypan blue and loaded onto a hemocytometer. Dead and live cells were counted using a light microscope, and cell viability as well as EC50 values were calculated.

The IC50<sub>12h</sub> of a chemical/toxicant was defined as the concentration that kills 50% of the lymphocytes after 12 hours of exposure. To determine this value for the compound under investigation, dose-response curves were plotted for five different CuO-NP concentrations (0.1, 0.5, 1, 2, 5 mM) after 12 hours of exposure, and the EC50 was determined based on the regression plot. The IC50<sub>12h</sub> for CuO-NPs was determined to be 0.4 mM. Further experiments were conducted according to a standard protocol by incubating CuO-NPs at concentrations of IC50/2 (0.2 mM), IC50 (0.4 mM), and 2  $\times$ IC50 (0.8 mM). These concentrations covered all levels of CuO-NP toxicity in human blood lymphocytes, ranging from sub-toxic (0.2 mM), threshold toxic (0.4 mM), to highly toxic (0.8 mM).

To evaluate lysosomal membrane leakage and other mechanistic parameters, time intervals of 2, 4, and 6 hours were selected to avoid highly toxic conditions (i.e., excessive cell death).

These intervals were chosen based on the prior determination of the IC50 (50% cell death) for CuO-NPs in human lymphocytes after 12 hours of exposure (Pourahmad et al., 2011).

# Assessment of Lysosomal Membrane Destabilization

The integrity of lysosomal membranes in human lymphocytes treated with CuO-NPs was assessed at 2, 4, and 6-hour intervals. For this purpose, a lipophilic dye was used, which accumulates in acidic organelles such as lysosomes. Upon lysosomal membrane damage, the dye leaks out of the organelle (Assadian et al., 2023).

 $100~\mu L$  of 5  $\mu M$  acridine orange was added to  $100~\mu L$  of the cell suspension and incubated at  $37^{\circ}C$  for 10~minutes. The lymphocytes were washed, and the fluorescence of released acridine orange was measured using a fluorescence spectrophotometer at excitation and emission wavelengths of 470~nm and 540~nm, respectively. Then, we subjected the sample to Faradarmani Consciousness Field to investigate its effects on the toxicity control of copper nanoparticles in human blood lymphocytes, considering the findings of our previous research on nanoparticle toxicity to analyze and interpret the results of this study (Taheri et al., 2022).

### **Statistical Analysis**

GraphPad Prism 10 software (GraphPad, La Jolla, CA) was used for data analysis. Statistical analysis of the data was performed using one-way and two-way ANOVA, followed by Tukey's and Bonferroni post-hoc tests. A minimum of three independent experiments were conducted. A p-value of less than 0.05 was considered statistically significant. Results are presented as mean ± standard error of the mean (SEM).

### **Results**

As shown in Figure 1, copper oxide nanoparticles (CuO-NPs) reduce the viability of human lymphocytes in a concentration-dependent manner over a 12-hour period, as assessed using the trypan blue exclusion dye method. A significant decrease in cell viability is observed at concentrations above 0.1 mM (P < 0.05), with highly significant effects at 0.5 mM and above (P < 0.001). The IC50<sub>12h</sub>, defined as the concentration that reduces viability by 50% after 12 hours of exposure, is determined to be 420 µM (0.42 mM). These findings highlight the cytotoxic potential of CuO-NPs, emphasizing their dose-dependent impact on lymphocyte survival.

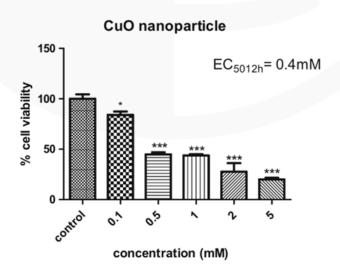


Figure. 1- Cell viability after treatment of human lymphocytes with CuO- NPs for 12 h. \*P < 0.05, \*\*P < 0.01, and \*\*\*P < 0.001



Figure 2 presents the significant effect of copper nanoparticles (CuO-NPs) on the survival of human lymphocytes and the effects of Faradarmani in reducing the cytotoxicity induced by these nanoparticles. CuO-NPs have been shown to exert dose- and time-dependent cytotoxic effects characterized by reduced cell survival, oxidative stress, and disruption of the lysosomal membrane. At higher concentrations and prolonged exposure times, these toxic effects become more pronounced, as observed in control samples, the highest concentration of CuO-NPs (0.8 mM) significantly induced lysosomal membrane leakage at all times. With the influence of FCF, the resistance of the lysosomal membrane increased at all concentrations, especially at 0.8 mM, resulting in a significant increase in viability (p-value< 0.05).

When comparing untreated control groups with FCF-exposed groups, a significant protective effect was observed in FCF-treated samples at all concentrations and time intervals. FCF

exposure resulted in higher cell survival rates, indicating that FCF effectively counteracted the harmful effects of CuO-NPs.

Treatment with FCF significantly improved cell survival in the presence of CuO nanoparticles (CuO-NPs), with notable differences observed at specific concentrations and time points. At 0.2 mM, FCF led to a significant increase in survival after 2 hours compared to the corresponding CuO-treated control (Con-0.2) (P < 0.05). Similarly, at 0.4 mM, a statistically significant enhancement was observed at 2 hours relative to Con-0.4 (P < 0.05). The most prominent effect was observed at the concentration of 0.8 mM, where the Faradarmani Consciousness Field improved cell viability by 140% at 2 hours, 103% at 4 hours, and 89% at 6 hours. The highest increase was observed at 2 hours. (P = 0.0008). These results highlight the strong protective capacity of FCF, particularly at higher concentrations and early exposure times, against CuO-NP-induced cytotoxicity.

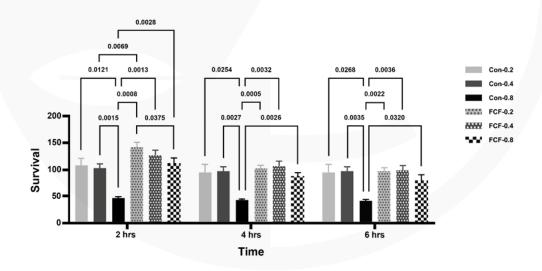


Figure 2. Lymphocyte survival rates after exposure to 0.2, 0.4, 0.8 concentrations of CuO nanoparticles (CuO-NPs) with and without Faradarmani Consciousness Field (FCF) over 2, 4, and 6 hours. Statistical significance (p-values) highlights the differences between conditions.

### Discussion

particularly Nanoparticles, copper oxide nanoparticles (CuO-NPs), have demonstrated significant cytotoxic potential, raising concerns regarding their safe application in biomedical, industrial, and environmental fields. In this study, we examined the impact of CuO-NPs on human lymphocyte survival and investigated whether Taheri Consciousness Fields (FCF) could mitigate their toxic effects. The control group was treated with CuO-NPs alone, while the experimental group was exposed to both CuO-NPs and FCF. The results provided compelling evidence that FCF significantly improve cell survival, suggesting a potential protective mechanism against nanoparticleinduced toxicity.

The toxic effects of CuO-NPs have been widely reported, with multiple studies demonstrating a dose-dependent decrease in cell viability across various human cell lines. Previous research has shown CuO-NPs reduce cell viability in HepG2 cells (48% at 10 µg/mL), A549 lung epithelial cells (93% at 20 µg/cm<sup>2</sup> and 50% at 15 µg/mL) (Brandão et al.,2020), HEp-2 airway epithelial cells (60% at 80 µg/cm<sup>2</sup>) (Farshori et al.,2022), SH-SY5Y neuroblastoma cells (60-70% at 0.01-10 µM) (Brandão et al.,2020). Similarly, our findings revealed a 50% reduction in lymphocyte viability at 420 µM CuO-NPs, consistent with these studies (Assadian et al., 2023). The cytotoxicity of CuO-NPs is largely attributed to oxidative stress, lysosomal membrane disruption, and DNA damage, leading to cellular dysfunction and apoptosis (Kubo et al., 2020).

The results in Figure 2 further strengthen the hypothesis that FCF enhance cellular resistance to CuO-NP-induced cytotoxicity. Over a 2, 4, and 6-hour period, lymphocyte survival was consistently higher in FCF-exposed groups compared to CuO-NP-treated controls. The statistically significant p-values indicate a clear protective effect, particularly at higher CuO-NP concentrations. At 2 hours, cell viability in control groups significantly declined, especially

at 0.8 mM CuO-NPs (p < 0.05), while FCF-treated samples exhibited notable resistance to cell death. This trend persisted at 4 and 6 hours, suggesting that FCF not only reduce immediate toxicity but may also facilitate long-term cellular adaptation and recovery.

A key observation was the stabilization of lysosomal membrane integrity in the presence of FCF. Given that CuO-NPs have been shown to cause lysosomal rupture, leading to the release of hydrolytic enzymes and subsequent cell death, our findings suggest that FCF may help maintain lysosomal integrity, thereby preventing premature cell death. These results align with previous research showing that Faradarmani and FCF mitigate environmental and biological stressors, including their ability to reduce oxidative stress in plants (Torabi et al., 2023) and decontaminate radioactive materials in aquatic environments (Taheri et al., 2022).

These findings offer new insights into the potential application of FCF as a protective treatment against nanoparticle-induced cytotoxicity. The observed increase in cell survival suggests that FCF may modulate key biological pathways involved in oxidative stress response, apoptosis regulation, and cellular repair mechanisms.

The exact molecular mechanisms underlying these effects remain unknown, warranting further exploration into mitochondrial function and its role in oxidative stress regulation, inflammatory responses triggered by CuO-NPs and the potential of FCF to modulate immune cell activity, and the long-term adaptation of cells to nanoparticle exposure in the presence of FCF.



Additionally, the integration of **FCF** nanotechnology with presents promising opportunities biomedical applications. in Given the increasing concerns surrounding nanoparticle safety, the use of FCF in drug delivery systems, regenerative medicine, and environmental decontamination could pave the way for safer and more efficient therapeutic strategies.

### **Conclusion**

In conclusion, this study suggests that FCF treatment may reduce oxidative stress, stabilize lysosomal membranes, and enhance cellular repair mechanisms, thereby improving resilience against nanoparticle-induced damage. We suggest that this research be expanded using diverse cellular models and toxicity mechanisms to validate its applicability in nanomedicine and toxicology.

### Acknowledgement

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# Faradarmani Consciousness Field Treatment Reduces MDA-MB-231 Cancer Cell Viability

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### **Abstract**

Breast cancer is the most common form of malignancy among women worldwide and is a multifactorial disease, the development of which involves various factors. The Faradarmani Consciousness Field (FCF) was introduced by Mohammad Ali Taheri as one of many Consciousness Fields that are neither energy nor matter, nor are they quantifiable; thus they cannot be directly observed or measured. However, it is possible to evaluate their effects indirectly through controlled experiments in the laboratory. This study aimed to investigate the effect of FCF on breast cancer cells (MDA-MB-231) measured using MTT and flow cytometry methods at 24, 48, and 72 hours. First, the MDA-MB-231 cell line was cultured, then the effect of FCF was studied at 24, 48, and 72 hours. To determine the cell death in the sample treated with FCF and compare it with the control, cells were stained with Annexin-V and propidium iodide (PI). Our results showed that the measured survival in the MTT test, at 24 hours, showed a 16% decrease in the experimental sample compared to the control. At this time, the percentage of early and late apoptosis and total apoptosis and necrosis in the sample under the influence of the FCF compared to the control cell lines increased by 5.92%, 3.49%, 9.41% and 4.68% respectively. Finally, the rate of programmed death of cancer cells increased in time intervals of 24 (9.41 %), 48 (21.44 %) and 72 (23.04 %) hours under the influence of this field in the examined cell line.

**Keywords:** Breast cancer, Taheri Consciousness Fields, Faradarmani, MTT assay, Cancer cells

### Introduction

Breast cancer is one of the most prevalent malignancies affecting women worldwide (Akorafas et al., 2002; Bener et al., 2017). According to the statistics of the World Health Organization, breast cancer accounts for about 30% of cancers among women. Approximately 1.2 million women are diagnosed with breast cancer annually. This type of cancer is reported to be the second leading cause of cancer-related deaths in women after lung cancer. It has been estimated that the prevalence of breast cancer will increase from two million patients in 2018 to more than three million patients in 2046, which represents an increase of 46% (Sung et al.,2021; Siegel et al., 2019). Furthermore, this type of cancer is known as one of the most costly diseases globally (Davari et al.,2013).

During recent decades, changes in lifestyle have contributed to an increased incidence and prevalence of breast cancer worldwide (Fredslund et al., 2013). Breast cancer is a disease in which malignant cells arise from breast tissue and proliferate uncontrollably. These cells typically originate from the epithelial cells lining the milk ducts or the lobules of the breast (Shah et al., 2014). Breast cancer is a multifactorial disease in which various risk factors are known to play a role. According to the studies conducted, these risk factors include family history, older age, age at menarche below 12 years, age at menopause after 54 years, first childbirth after age 30, nulliparity, high mammographic density, elevated sex hormone levels, childhood exposure to ionizing radiation, race, socioeconomic status, body mass index, and lifestyle factors such as diet, physical activity, smoking, and alcohol consumption (Moller et al., 2003; Kaminska et al., 2015; Sun et al., 2017).

The MDA-MB-231 cell line is derived from human breast cancer, and it has a spindle-shaped epithelial morphology. These undifferentiated cells are very aggressive and lack estrogen and progesterone receptors, as well as the human epidermal growth factor receptor 2 (HER2).

Therefore, this cell line is classified as triplenegative breast cancer (TNBC). The invasive power of this cell line is mediated through the ability to proteolytically alter the extracellular matrix (Chavez et al., 2010; Lukasiewicz et al., 2021).

The nature of consciousness and its place in science has received much attention in the current century. Many philosophical and scientific theories have been proposed in this area. In the1980s, Mohammad Ali Taheri introduced novel fields with non-material/non-energetic nature (Taheri, 2013) named Taheri Consciousness Fields (TCFs).

In previous studies, the effects of TCFs on various cancer cell lines, as well as on animal and plant models, have been investigated (Taheri et al., 2020; Taheri et al., 2022; Torabi et al., 2023; Taheri et al., 2024). The aim of this study was to investigate the effect of the Faradarmani Consciousness Field on breast cancer cells (MDA-MB-231) using MTT and flow cytometry methods.

### **Materials and Methods**

# **Application of Faradarmani Consciousness Field**

TCFs were applied to the samples according to the protocols regulated by the COSMOintel research center (www.COSMOintel.com). A detailed explanation is provided in the general discussion of this issue. In this study, MDA-MB-231 cells were exposed to the Faradarmani Consciousness Field once at 24, 48, and 72 hours from the start of the study. MDA-MB-231 cells that were not exposed to the FCF were considered the control group.

### Cell culture

In this research, the MDA-MB-231 breast cancer cell line, obtained from the cell bank of the Pasteur Institute of Iran, was used. The cells were cultured in DMEM (Dulbecco's Modified Eagle Medium) (Gibco, USA) supplemented



with 2 mM L-glutamine enriched, fetal bovine serum (FBS) (Gibco, USA) and 1% Penicillin/streptomycin antibiotic solution (Biosera, France) under controlled conditions of 37°C and 5% CO<sub>2</sub>. The cells grew as a monolayer in the flask. This culture medium was changed three times a week, and sterile trypsin-EDTA solution was used to harvest the cells.

# Evaluation of cytotoxicity effect of FCF by MTT assay

The MTT test is a quantitative and colorimetric assay based on the reduction of a yellow, water-soluble salt (3-(4, 5-dimethylthiazol-2yl) -2, 5-diphenyl tetrazolium bromide) (MTT) (Sigma-Aldrich, Germany) and the formation of dark blue and insoluble formazan crystals in water. The regeneration of MTT occurs by the mitochondrial enzyme succinate dehydrogenase and only in living cells. Formazan crystals are soluble in organic solvents such as isopropanol and DMSO (Merck, Darmstadt, Germany), and by measuring their light absorption, the metabolic activity level of living cells can be determined (Ghasemi et al., 2021). The procedure was carried out in such a way that 104 cells were cultured in each well of a 96well plate and after 24 hours and upon reaching approximately 80% confluency, the supernatant was exposed to FCF at time intervals of 24, 48, and 72 hours. Other culture plates were used as controls for the MTT test at intervals of 24, 48, and 72 hours. After the desired times passed, the plates were removed from the incubator, the supernatant of the wells was removed and 100 microliters of DMEM medium without alpha naphthol along with 10 microliters of MTT (5 mg/mL) was added to each well. Then the plates were again transferred to the incubator and kept at 37°C for 3 to 4 hours. In the next step, the produced formazan product was dissolved by adding 50 microliters of dimethyl sulfoxide (DMSO) solvent, and color intensity was measured by an ELISA reader at a wavelength of 570 nm. The biological capacity of cells treated with FCF was determined as the ratio of percentage of absorbance in comparison to the absorbance of formazan in the control group.

### Evaluation of apoptosis by flow cytometry

To determine the percentage of apoptotic cells in a cell population treated with FCF and compare it with the control cell population, cells were stained with Annexin-V and propidium iodide (PI) (Sigma-Aldrich, Germany). After treatment of the cells with FCF at 24, 48, and 72 hours, the cells were trypsinized and washed with sterile phosphate-buffered saline (PBS). 100 microliters of binding buffer were added to the sediment resulting from the centrifugation of the cells in a 1.5 ml microtube. Next, 10 microliters of PI and 5 microliters of Annexin-V were added to the tube. The contents were gently mixed by manually shaking the microtube to ensure the cell pellet was fully resuspended. In the next step, the samples were incubated at room temperature (25°C) for ten minutes in the darkness. Finally, cell analysis was performed by flow cytometry (BD Biosciences, San Diego, CA, USA). Data analysis was done by using the software of the device and dividing the points recorded in the two-dimensional curve into four regions Q1 to Q4. To assess the effect of FCF on apoptosis and necrosis, the percentage of cells in each quadrant was calculated and reported using the flow cytometry analysis software (FCS Express).

### Statistical analysis

GraphPad Prism 9 and SPSS version 2016 were used for statistical calculations. The collected data were analyzed using one-way ANOVA followed by Tukey's post hoc test. Assays were repeated three times. A p-value of less than 0.05 was considered statistically significant.

### Results

# Examining the effect of FCF on cell survival using MTT assay

According to Figure 1, the only significant change was observed at 24 hours, with a 16% decrease in survival in the samples treated with FCF.

### Cell Survival rate (%)

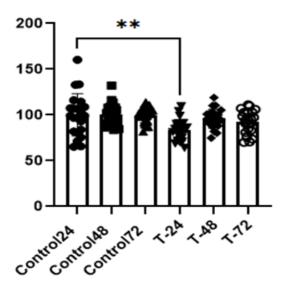


Figure.1. Survival changes based on the MTT test in this study; \*\* represents p-value=0.002. (T: Faradarmani Consciousness Field treatment). \*\*: p-value< 0.01.

# Assessment of the effect of FCF on apoptosis using flow cytometry

Figure 2 illustrates the apoptotic cells in three time periods of 24, 48, and 72 hours for both the treatment and control groups. As shown, the percentage of live cells (Q4 region) in the control group was 90.7%, 92.6%, and 74.6% at 24, 48, and 72 hours, respectively. In contrast, in the FCF-treated group, the corresponding percentages of live cells were 76.6%, 69%, and 49.6%, respectively.

The percentage of early apoptotic cells (Q3 region)—i.e., cells in the early stages of apoptosis— increased from 10.6% at 24-hour treatment period, and 20.2% at 48-hours to 33% at 72-hour. The percentage of late apoptotic or necrotic cells (Q2 area)—i.e., cells in the final stages of apoptosis—also increased with longer FCF exposure, rising from 4.42% at 24 hours to 6.69% at 48 hours and reaching 8.47% at 72 hours. Additionally, the percentage of necrotic cells (Q1 region) in the control group was 3.72%, 1.94%, and 6.91% at 24, 48, and 72 hours, respectively. In contrast, the corresponding percentages in the FCF-treated group were 8.40%, 4.07%, and 8.92%, respectively.

As shown in Figure 2, with the increase in the duration of the FCF effect, the lowest percentage of cells in the Q4 region was observed in the FCF-treated group at 72 hours, indicating the apoptotic effect of FCF treatment. According to the results of Table 1, a statistically significant difference was observed between the control group and the group under the effect of FCF treatment (P<0.001). In other analyzed hours, cases of statistically significant correlation were also seen. For example, based on the results of Figure 3, there was a statistically significant difference between the number of apoptotic and live cells in the treatment and control groups at 48 and 72-hour intervals under the effect of FCF (P < 0.01). The results showed that the percentage of early and late apoptosis and total apoptosis and were higher in FCF-treated cell lines compared to the control (Figure 3).

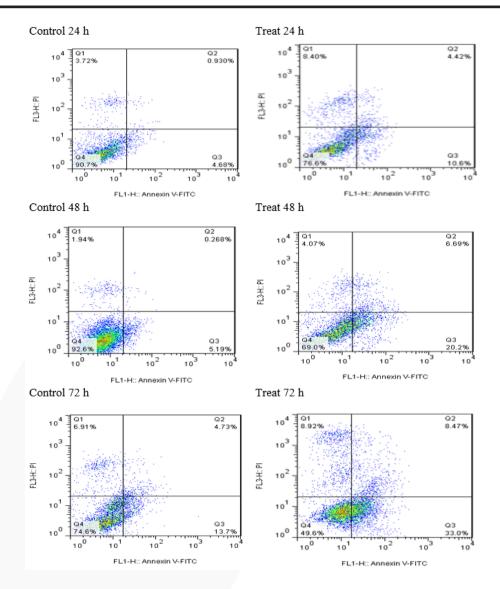


Figure 2: The effect of Faradarmani Consciousness Field (FCF) at time intervals of 24, 48 and 72 hours on apoptosis in MDA-MB-231 cells. Control group (cells without FCF treatment).

Q1: Necrotic cells (Annexin-V negative and PI positive), Q2: late apoptotic or necrotic cells (Annexin-V and PI positive), Q3: healthy cells: (Annexin-V and PI negative), Q4: early apoptotic cells (Annexin-V positive and PI negative)

Table 1. Percentage of cells at different stages of necrosis and apoptosis based on flow cytometry analysis, comparing the control and test groups treated with the Faradarmani Consciousness Field (FCF) in MDA-MB-231 cells over a 24-hour period.

	Q1	Q2	Q3	Q2+Q3	Q4
Control (-)	3.72%	0.93%	4.68%	5.61%	90.7%
FCF	8.40%	4.42%	10.6%	15.02%	76.6%
Difference of FCF from negative control	4.68%	3.49%	5.92%	9.41%	14.1%

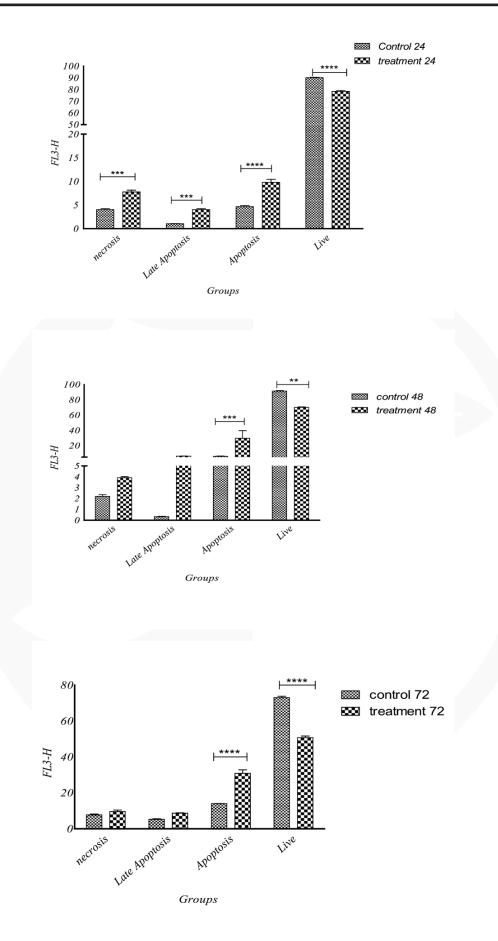


Figure 3. Chart showing changes in cell death at 24-, 48-, and 72-hour intervals in control and FCF-treated groups based on flow cytometry analysis. \*\*\*\*: Statistically significant (p<0.0001), \*\*\*, \*\*: Statistically significant (p<0.05)



### Discussion and conclusion

In recent years, the effects of TCFs have been investigated in a wide range of experiments. According to Taheri, these fields are neither matter nor energy, and it is not possible to measure them with conventional scientific tools, but it is possible to examine their effects on living organisms or non-living materials. One of the advantages of utilizing TCFs in the treatment of diseases like breast cancer is their drug-free approach which eliminates the risk of side effects and the need for any materials, equipment, or instruments to obtain the results. In addition, due to the nature of this filed, the application of TCFs is free of charge.

In the present study, the effect of Faradarmani Consciousness Field (FCF) on breast cancer cell line (MDA-MB-231) was investigated using MTT assay at 24, 48, and 72 hours. To determine the percentage of apoptotic and necrotic cells in a cell population treated with FCF and compare it to the cell population in the control group, cells were stained with Annexin-V and propidium iodide (PI). The results of MTT test showed that FCF caused a decrease in survival in the MDA-MB-231 cell line at 24 hours. These findings were confirmed by flow cytometry analysis. Flow cytometry analysis showed that a significant percentage of cells underwent programmed cell death (apoptosis) as a result of the treatment with FCF. The level of necrosis in treated cells was not significant compared to the percentage of apoptotic cells. There was a statistically significant relationship between the number of apoptotic cells and the number of live cells in both treatment and control groups at the 48- and 72-hour time points.

According to Taheri, the application of FCF is effective in repairing and modifying the system under study, restoring it to its optimal condition. From the perspective of Interuniversalism (Farakolnegari), proposed by Taheri, human existence consists of numerous hardware and software components. From this perspective, the part of the human being that forms the body is considered the "hardware" component

that is operated by a large system of multiple software programs (the Operating System). In other words, behind the scenes of this hardware, there are a multitude of software programs that manage the physical body (Taheri 2011).

conventional intervention methods, interventions affect the hardware. For example, in the treatment of various types of cancer, including breast cancer, a range of methods is used, such as surgery, radiotherapy and chemotherapy to specific and targeted therapies. Due to the biological and genetic differences in individuals, the conventional treatments in these patients have different responses and may be unsuccessful and carry a high risk of recurrence (Wayteck et al., 2014). But the application of Faradarmani Consciousness Field is a noninvasive method where the effects occur at the level of the software or infrastructure of the system under study, meaning FCF changes the behavior of the hardware by modifying the software (that runs the hardware) behind the scenes (Taheri et al., 2013). Previous studies have reported varying behaviors among different cell lines, suggesting that FCF exerts cell-specific effects (Taheri et al., 2023).

In conclusion, the findings of this study on the MDA-MB 231 cell line indicate that FCF can reduce cancer cell viability and promote programmed cell death (apoptosis). As a next step, we suggest that in future studies, the effect of FCF on the level of expression of apoptosisinducing molecules such as Fas (CD95) be investigated. Further research is required to clarify the precise mechanisms through which FCF exerts its effects in biological systems.

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# Behavior of Normal and Cancerous Human Cell Lines Exposed to T-Consciousness Fields during Stress Conditions

According to the Theory of T-Consciousness, when a subject under study is influenced by T-Consciousness Fields, the information transmitted from these fields leads to changes in its behavior and characteristics. A unique feature of this theory is its testability under laboratory conditions. In the research presented in this issue, the alleviative effects of these non-physical fields on cells under microgravity stress and nanoparticle toxicity have been investigated. Additionally, the viability of a breast cancer cell line under the influence of these fields has also been evaluated.

