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Effect of Biofield Energy Treated Cell Growth Medium on Human Fibroblast Cell Growth for Assessment of Skin Health Promotion Potential

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Abstract

Cell growth and proliferation plays an important role in wound healing, which is a complex and dynamic process. Wound management and recovery is currently done using various antiseptics and antibiotics in order to prevent or treat infection. The objective of the present study was to assess the effect of Biofield Energy (Consciousness Energy Healing Treatment - The Trivedi Effect®) Treated cell growth medium on the proliferation of human dermal fibroblast cells (HFF-1) for the assessment of skin health promotion potential. An aliquot of HFF-1 cells and DMEM were divided into two parts, one was treated with the Biofield Energy Treatment and denoted as the treated test samples, while other part was coded as the untreated test samples. MTT assay was used to study the cellular proliferation, while cell growth rate was estimated under phase contrast microscope by monitoring the cell growth at 24, 48, and 72 hours interval and photomicrographs were captured. The results of HFF-1 cellular proliferation rate showed a significant ($p \leq 0.001$) increased by 80% in the Biofield Energy Treated DMEM, while positive controls, ascorbic acid (10 µM) and FBS (15% fetal bovine serum) showed increased cell proliferation by 21% and 44%, respectively. In addition, cell growth rate was evaluated at 24, 48, and 72 hours and the results were observed with a significant growth in the Biofield Energy Treated DMEM group compared with the Biofield Energy Treated HFF-1 cells. Thus, on the basis of the experimental results, it can be concluded that The Trivedi Effect®-Consciousness Energy Healing Treatment has the significant capacity to improve the cellular proliferation and cell growth rate of human skin cells in the Biofield Energy Treated media (DMEM) for improving the overall skin health. It can be used as a complementary and alternative approach with respect to the various skin-related disorders, anti-aging, wound healing, and many more.

Keywords: Anti-aging; HFF-1 cell line; The Trivedi Effect®; Consciousness Energy Healing Treatment; Biofield Energy Healing

Abbreviations: DMEM: Dulbecco's Modified Eagle Medium; FBS: fetal bovine serum; CAM: Complementary and Alternative Medicine; EDTA: Ethylenediaminetetraacetic acid; DMSO: Dimethyl sulphoxide; NCCAM: National Center for Complementary and Alternative Medicine.

Introduction

Skin cell proliferation and cell growth plays an important roles in normal and pathological skin healing. In addition to, skin aging and its signs follows various trajectories in organs, tissues and cells with time. Cellular proliferation and its growth are influenced by many endogenous (intrinsic) and exogenous (extrinsic) factors. Intrinsic factors included such as individual genetics, cellular metabolism, hormone and metabolic processes, while extrinsic factors includes chronic light exposure, pollution, ionizing radiation, chemicals, and toxins [1]. The combination of the above factors will decide the cumulative structural and physiological skin alterations and progressive changes as well as the color changes [2]. Gradual loss of skin cell growth leads to premature photoaged skin, thin and atrophic, mottled discoloration, deep wrinkles, laxity, dullness and roughness, which leads to loss in skin elasticity known as skin sagging [3,4]. Epidermal turnover rate had reported to be decrease which would increase the process of aging and slower the process of wound healing. In order to minimize the gradual loss and to improve the growth of skin cells, several chemical targets have been discovered with wide application to improve the rate of cell growth and cycle, which will leads to improve the skin appearance and will speed the rate of wound healing. However, various experimental studies concluded that these chemical based methods would not ensure a natural skin appearance, and might have several side effects with respect to skin anatomy and physiology [5]. The Complementary and Alternative Medicine (CAM) systems are accepted worldwide and used for many therapies related to the skin and wound management [6].

The science of healing is now considered as an emerging field in medicine against wide range of clinical symptoms. The use of alternative medicines such as Biofield Energy Healing has been reported to be increase in order to promote wellness by uncovering the root cause of diseases with universal solutions [7]. Most of the U.S. population has been accepting the significance of Complementary and Alternate Medicine (CAM) [8]. The National Center for Complementary and Alternative Medicine (NCCAM), now defined Biofield Therapies as complementary medicine domain. Many clinical outcomes have been reported from Biofield Energy Healing Treatment by renowned healers that were accepted world-wide [9-11]. Consciousness Energy Healing Treatment (The Trivedi Effect®) has been reported with noteworthy results in nonliving materials and living organisms. The momentous results of The Trivedi Effect® have been published in the field of microbiology [12-14], agriculture science [15-17], livestock [18], nutraceuticals [19,20], and materials science [21-23]. On this basis, the present study was designed to evaluate the effect of Biofield Energy Treatment (The Trivedi Effect®) on HFF-1 cell line and DMEM for evaluation of cellular proliferation rate using MTT assay and cell growth assay using phase contrast microscopy.

Materials and Methods

Chemicals and Reagents

Dulbecco's Modified Eagle Medium (DMEM) and fetal bovine serum (FBS) were purchased from Gibco, Genex Life Sciences Pvt. Ltd., India. Ethylenediaminetetraacetic acid (EDTA), trypsin, L-ascorbic acid, and $NaHCO_3$ were purchased from Sigma, USA. Antibiotics solution (Penicillin-Streptomycin) was procured from HiMedia Pvt. Ltd., USA. Dimethyl sulphoxide (DMSO) was obtained from Thermo Fisher Scientific, USA. All the other chemicals used in this experiment were analytical grade procured from India.

Cell Culture Maintenance (HFF-1, ATCC® SCRC-1041™)

HFF-1 (human foreskin fibroblast) cells were procured from American Type Culture Collection (ATCC), SCRC-1041™, USA, originated from normal human skin fibroblast cells. HFF-1 cell line was maintained in the growth medium DMEM supplemented with 15% FBS, with added antibiotics penicillin (100 U/mL) and streptomycin (100 µg/mL). The growth condition of cell lines were 37°C, 5% CO_2, and 95% humidity. The cells were sub-cultured by trypsinisation followed by splitting the cell suspension into fresh flasks and supplementing with fresh cell growth medium. L-ascorbic acid and FBS (positive control) were diluted in DMEM to achieve the working concentration corresponding in cell plate.

Biofield Energy Healing Strategy

An aliquot of HFF-1 cells in a T-25 cell culture flask and an aliquot of DMEM culture medium were received

the Biofield Energy Treatment (The Trivedi Effect®) under standard laboratory conditions. This Biofield Energy Healing Treatment was provided by a renowned Biofield Energy Healer, Mr. Mahendra Kumar Trivedi, who participated in this study and performed the Biofield Energy Treatment for ~3 minutes from a distance of ~25 cm. The energy transmission was done without touching the cells and DMEM. This Biofield Energy Treatment was administered through the Healer's unique Energy Transmission process to the HFF-1 cells and DMEM under laboratory conditions. After that, the Biofield Energy treated and untreated samples were kept in similar sealed conditions for experimental study. Following Biofield Energy Treatment, the medium and the cell line were used for the estimation of cell growth and proliferation assay. The Biofield Energy Treated and untreated T-25 flask were incubated till one week in a CO_2 incubator at 37°C, 5% CO_2, and 95% humidity. Besides, the Biofield Energy Treated and untreated DMEM were stored at 4ºC till cell culture [19].

Experimental Design

Group I served as the untreated cells in the untreated medium (200 µL of phenol-free DMEM supplemented with 10% CD-FBS). Group II served as a positive control (L-ascorbic acid), i.e. cells in DMEM with ascorbic acid (10 and 50 µM) in cellular proliferation, while 15% FBS was used as another positive control in proliferation assay. Group III was referred as the untreated HFF-1 cells in the Biofield Energy Treated DMEM. Group IV was served as the Biofield Energy Treated HFF-1 cells in the untreated DMEM.

Assessment on Cellular Proliferation Assay

The Biofield Energy Treated HFF-1 cells were trypsinized, counted and plated at density of 5 X 10^3 cells/well/180 µL of growth medium followed by overnight incubation for cell recovery and exponential growth. Further, the cells were subjected to serum starvation so as to synchronize cell growth. These cells were treated as per experimental procedure with positive controls (ascorbic acid and FBS) and test items (Biofield Energy Treated cells and DMEM) followed by the incubation for 72 hours in a CO_2 incubation at 37 °C, 5% CO_2, and 95% humidity. About 20 µL of 5 mg/mL of MTT 3-(4,5-dimethythiazol-2-yl)-2,5-diphenyl tetrazolium bromide solution was added followed by additional incubation for 3 hours at 37°C. The supernatant was aspirated and 150 µL of DMSO was added to each well to dissolve the formazan crystals followed by measurement of absorbance at 540 nm using Synergy HT microplate reader [24].

Estimation of Cell Growth in the Biofield Energy Treated DMEM

HFF-1 cells were trypsinized at the density corresponding to 5 X 10^3 cells/well/180 µL of growth medium followed by incubation. Further, the cells were subjected to the treatment with the Biofield Energy Treated and untreated DMEM. After treatment, the plates were kept in standard condition, which were visualized under phase contrast microscope for monitoring the cell growth at three time points, 24, 48, and 72 hours [25].

Statistical Analysis

Each experiment was carried out in three independent assays and were expressed as mean values ± Standard Deviation (SD). For statistical comparison, values were subjected to one-way analysis of variance (ANOVA) with Bonferroni post-test using Graphpad prism software version 4.01. Statistical significance was considered at $P < 0.05$.

Results and Discussion

Cellular Proliferation Assay

After 72 hours of incubation, positive control L-ascorbic acid (10 and 50 µM) and 15% FBS showed as significantly increased the cell growth in HFF-1 cells compared with the control group as determined by cellular proliferation assay (Table 1). The results of absorbance in different experimental groups are presented in Table 1. The positive control (10 µM, L-Ascorbic acid) and 15% FBS showed a significant ($P \leq 0.001$) absorbance value as 0.409 ± 0.00 and 0.453 ± 0.03 at 540 nm. Similarly, the Biofield Energy Treated DMEM showed a significant ($P \leq 0.001$) change in absorbance i.e. 0.536 ± 0.01, which suggest a significant increased cellular proliferation rate. The percentage of cellular proliferation was calculated with respect to the baseline control group value. The data suggested that the Biofield Energy Treated DMEM showed a significant ($P \leq 0.001$) increased proliferation by more than 80% compared with the baseline control group. However, Biofield Energy Treated cells showed approximately 2% increase compared with the baseline control group. Hence, overall data concluded that the Biofield Energy Treated (The Trivedi Effect®) DMEM would better to

improve the cellular proliferation rate compared with the Biofield Energy Treated HFF-1 cell.

Group	Description	Absorbance (540 nm)
Baseline (0 hour)	Cells + DMEM	0.168 ± 0.01
Baseline (72 hours)	Cells + DMEM	0.367 ± 0.01
	L-Ascorbic acid (10 µM)	0.409 ± 0.00***
Positive control	L-Ascorbic acid (50 µM)	0.399 ± 0.01***
	FBS (15%)	0.453 ± 0.03***
Test Item	Biofield Energy Treated DMEM	0.536 ± 0.01***
	Biofield Energy Treated HFF-1 Cells	0.370 ± 0.01***

Table 1: Effect of the Biofield Energy Treated DMEM and HFF-1 cells on cellular proliferation.

Each value represented as mean ± SD of triplicate wells. Statistical comparison was performed using One-way analysis of variance (ANOVA) with Bonferroni post-tests (Graphpad prism software version 4.01). ***$P \leq 0.001$ with respect to baseline at t=0 hour. FBS: Fetal bovine serum.

These findings suggested that the cellular proliferation rate after the Biofield Energy Treatment was significantly increased. The significant enhanced cell proliferation and migration were reported in the Biofield Energy Treated DMEM compared with the Biofield Energy Treated HFF-1 cells. Cell proliferation and migration are the important parameters, which can be utilized in skin health in terms of wound healing, skin regeneration potential, antiaging, etc. [26].

Cell Growth Rate

Dermal human fibroblast cells (HFF-1) grown in the Biofield Energy Treated DMEM demonstrated greater confluence indicating higher cell number compared to cells grown in untreated medium (DMEM) at all the three-time points. Represented images of the cells in the presence of the Biofield Energy Treated and untreated DMEM are shown in Figure 1. The results suggested that the number of cells were increased at 48 and 72 hours as compared with 24 hours interval in the presence of untreated DMEM (Figure 1a-1d). Similar pattern of growth was observed in the Biofield Energy Treated DMEM, which suggest a significant increased number of cells at 48 hours (Figure 1e) and 72 hours (Figure 1f). This suggests that Mr. Trivedi's Biofield Energy Treatment has the significant capacity to improve the cellular proliferation and cell growth, which might be useful as a skin health or would work as antiaging therapy [27].

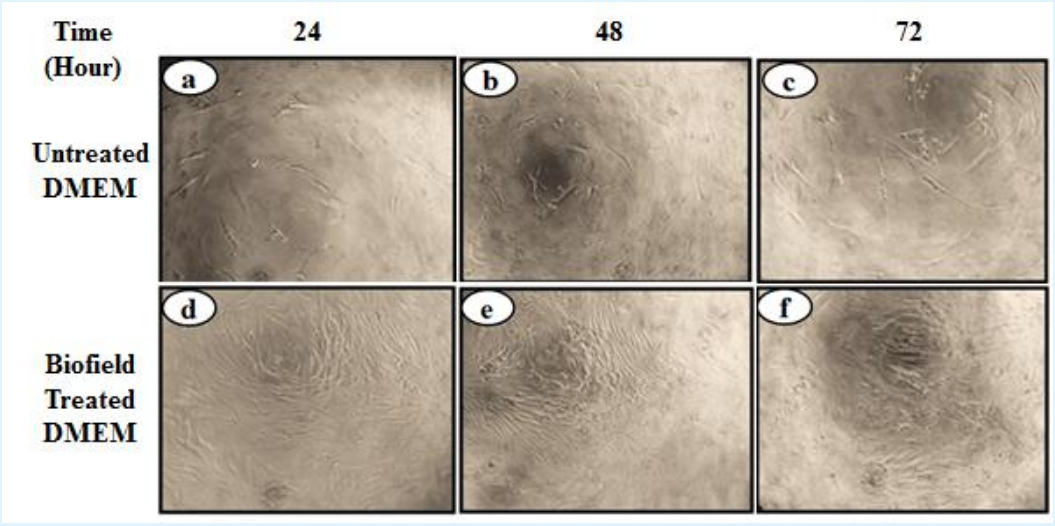

Figure 1: Representative photomicrograph of HFF-1 cells growth in the presence of untreated DMEM (control) at (a) 24 hours; (b) 48 hours; and (c) 72 hours; and in the presence of Biofield Energy Treated DMEM at (d) 24 hours; (e) 48 hours; and (f) 72 hours.

According to the report of Golberg, et al., some specific pulsed low intensity electric field parameters were tested in rats, which results in prominent proliferation of the epidermis, microvasculature formation, and synthesis of extra cellular matrix components [28]. Biofield Energy Treatment, an energetic matrix surrounds the human body, also known as complex and dynamic energy field. It might be expected that Biofield Energy Treatment might stop the partial cell necrosis due to apoptosis, cell membrane electroporation, control the oxidative damage to the skin membrane, maintain local pH changes, or by some other mechanisms and results in improved cell proliferation and cell growth rate. It can be assumed that The Trivedi Effect®- Consciousness Energy Healing Treatment can be used as a potent anti-aging or anti-fibrotic agent to promote the skin health.

Conclusion

The results of cellular growth and proliferation data showed a significant improvement in the cell growth that suggest the significance of Biofield Energy Treatment (Consciousness Energy Healing Treatment - The Trivedi Effect®) in the HFF-1 cell line and DMEM for skin health. Biofield Energy Healing might be used as an alternative method in order to improve the overall skin health and aging. The present study showed significant results in in vitro skin health models with respect to HFF-1 cells. The cellular proliferation rate using MTT assay showed a significantly ($P \leq 0.001$) improved by more than 80% after treatment with the Biofield Energy Healing (The Trivedi Effect®) Treatment on DMEM and HFF-1 cells. In addition to, the cell growth rate was significantly improved in the Biofield Energy Treated DMEM group at 24, 48, and 72 hours study period as compared with the baseline control group. However, the Biofield Energy Treated HFF-1 cells did not show any significant change in proliferation rate. Overall, the data concluded that the Biofield Energy Treatment (The Trivedi Effect®) has a significant potential with respect to the wound healing, antiaging, and skin regeneration activities. Overall, The Trivedi Effect® can be used as a Complementary and Alternative Medicine (CAM) against skin irregularities that are typically symptoms of a skin disorders such as eczema, diaper rash, chickenpox, measles, warts, acne, hives, wrinkles, ringworm, seborrheic dermatitis, skin cancer, rashes from bacterial or fungal infections, rashes from allergic reactions, raised bumps that are red or white, cracked skin, discolored patches of skin, fleshy bumps, warts, or other skin growths, changes in mole color or size, a loss of skin pigment, scaly or rough skin, peeling skin, ulcers, open sores or lesions, dry, excessive flushing, etc.

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References

1. Cevenini E, Invidia L, Lescai F, Salvioli S, Tieri P, et al. (2008) Human models of aging and longevity. Expert Opin Biol Ther 8: 1393-1405.

2. Fisher GJ (2005) The pathophysiology of photoaging of the skin. Cutis 75: 5-8.

3. Escoffier C, de Rigal J, Rochefort A, Vasselet R, Lévêque JL, et al. (1989) Age-related mechanical properties of human skin: An *in vivo* study. J Invest Dermatol 93(3): 353-357.

4. Schmuth M, Watson RE, Deplewski D, Dubrac S, Zouboulis CC, et al. (2007) Nuclear hormone receptors in human skin. Horm Metab Res 39: 96-105.

5. Baumann L (2007) Skin ageing and its treatment. J Pathol 211: 241-251.

6. Dorai AA (2012) Wound care with traditional, complementary and alternative medicine. Indian J Plast Surg 45: 418-424.

7. Rubik B, Muehsam D, Hammerschlag R, Jain S (2015) Biofield science and healing: History, terminology, and concepts. Glob Adv Health Med 4: 8-14.

8. Barnes PM, Bloom B, Nahin RL (2008) Complementary and alternative medicine use among adults and children: United States, 2007. Natl Health Stat Report 12: 1-23.

9. Jain S, Ives J, Jonas W, Hammerschlag R, Muehsam D, et al. (2015) Biofield science and healing: An emerging frontier in medicine. Glob Adv Health Med 4: 5-7.

10. Trivedi MK, Mohan TRR (2016) Biofield energy signals, energy transmission and neutrinos. American Journal of Modern Physics 5(6): 172-176.

11. Schlitz M, Hopf HW, Eskenazi L, Vieten C, Radin D (2012) Distant healing of surgical wounds: An exploratory study. Explore (NY) 8(4): 223-230.

12. Trivedi MK, Branton A, Trivedi D, Nayak G, Shettigar H, et al. (2015) Antibiogram pattern of *Shigella flexneri*: Effect of biofield treatment. Air Water Borne Diseases 3: 122.

13. Trivedi MK, Patil S, Shettigar H, Mondal SC, Jana S (2015) Antimicrobial susceptibility pattern and biochemical characteristics of *Staphylococcus aureus*: Impact of biofield treatment. J Microb Biochem Technol 7: 238-241.

14. Trivedi MK, Branton A, Trivedi D, Nayak G, Shettigar H, et al. (2015) Effect of biofield energy treatment on *Streptococcus* group B: A postpartum pathogen. J Microb Biochem Technol 7: 269-273.

15. Trivedi MK, Branton A, Trivedi D, Nayak G, Gangwar M, et al. (2015) Morphological and molecular analysis using RAPD in biofield treated sponge and bitter gourd. American Journal of Agriculture and Forestry 3: 264-270.

16. Trivedi MK, Branton A, Trivedi D, Nayak G, Gangwar M, et al. (2015) Effect of biofield energy treatment on chlorophyll content, pathological study, and molecular analysis of cashew plant (*Anacardium occidentale* L.). Journal of Plant Sciences 3(6): 372-382.

17. Trivedi MK, Branton A, Trivedi D, Nayak G, Gangwar M, et al. (2016) Molecular analysis of biofield treated eggplant and watermelon crops. Adv Crop Sci Tech 4: 208.

18. Trivedi MK, Branton A, Trivedi D, Nayak G, Mondal SC, et al. (2015) Effect of biofield treated energized water on the growth and health status in chicken (*Gallus gallus domesticus*). Poult Fish Wildl Sci 3: 140.

19. Trivedi MK, Branton A, Trivedi D, Nayak G, Wellborn BD, et al. (2016) Effect of biofield energy healing based herbomineral formulation on pro-inflammatory cytokines expression in biofield treated mouse splenocyte cells: Impact of the Trivedi Effect®. American Journal of Bio Science 4: 74-83.

20. Trivedi MK, Branton A, Trivedi D, Nayak G, Lee AC, et al. (2017) Investigation of physicochemical, spectral, and thermal properties of sodium selenate treated with the energy of consciousness (The Trivedi Effect®). American Journal of Life Sciences 5: 27-37.

21. Trivedi MK, Nayak G, Patil S, Tallapragada RM, Latiyal O, et al. (2015) An evaluation of biofield treatment on thermal, physical and structural properties of cadmium powder. J Thermodyn Catal 6: 147.

22. Trivedi MK, Nayak G, Patil S, Tallapragada RM, Latiyal O, et al. (2015) Effect of Biofield energy treatment on physical and structural properties of calcium carbide and praseodymium oxide. International Journal of Materials Science and Applications 4(6): 390-395.

23. Trivedi MK, Tallapragada RM, Branton A, Trivedi D, Nayak G, et al. (2015) Characterization of physical, thermal and structural properties of chromium (VI) oxide powder: Impact of biofield treatment. J Powder Metall Min 4: 128.

24. Seo Y, Lee HJ, Kim Y, Youn H, Lee BJ (2005) Effects of several salt marsh plants on mouse spleen and thymus cell proliferation using MTT assay. Ocean Science Journal 40(4): 209-212.

25. Jaccard N, Griffin LD, Keser A, Macown RJ, Super A, et al. (2014) Automated method for the rapid and precise estimation of adherent cell culture characteristics from phase contrast microscopy images. Biotechnol Bioeng 111(3): 504-517.

26. Pastar I, Stojadinovic O, Yin NC, Ramirez H, Nusbaum AG, et al. (2014) Epithelialization in wound healing: A comprehensive review. Adv Wound Care (New Rochelle) 3(7): 445-464.

27. Shiraha H, Gupta K, Drabik KA, Wells A (2000) Aging fibroblasts present reduced epidermal growth factor (EGF) responsiveness due to preferential loss of EGF receptors. J Biol Chem 275(25): 19343-19351.

28. Golberg A, Khan S, Belov V, Quinn KP, Albadawi H, et al. (2015) Skin rejuvenation with non-invasive pulsed electric fields. Sci Rep 5: 10187.