Effect of the Faradarmani Consciousness Field on Mutant Strain of SARS-CoV-2

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**Dr. Laleh Amani was an outstanding, compassionate, and enthusiastic researcher in the Cosmointel. Inc studies who passed away in 2021. We extend our sincere condolences and appreciation for her extraordi-nary efforts in this research and pray for her peace.

ABSTRACT

The COVID-19 pandemic is still a threat to global health. In late 2020, the rapid emergence of SARS-CoV-2 mutant strains was reported, raising concerns in the prevention and treatment of COVID-19. SARS-CoV-2 that encoded a D614G mutation in the viral spike (S) protein predominated globally and this change increased virus transmission. Furthermore, the false-negative results from respiratory specimens are one of the problems in the detection of COVID-19. Taheri Consciousness Fields (TCFs) introduced by Mohammad Ali Taheri, are novel fields that are neither matter nor energy. Therefore, they are non-quantifiable and cannot be directly observed or measured. However, it is possible to demonstrate and measure the effects of these fields through standard scientific experiments. The present study aimed to evaluate the effect of Faradarmani CF as one of the TCFs on mutant D614G virus titer and RNA copy number and the quality of amplification of coronavirus genome fragments. The results of this study showed that the Faradarmani CF increased replication of non-mutated and mutated viruses compared with control samples and increased the replication fitness. The Faradarmani CF increased the rate of replication of the mutant strain close to the non-mutant strain in the cells, and in fact, reached a biological equilibrium of compatibility similar to that of the Wuhan strain with the cell. In addition, the PCR products of coronavirus genome fragments under the influence of the Faradarmani CF had sharper bonds on gel electrophoresis with more quality of amplification compared with the control. This result suggests that Faradarmani CF can be used in diagnostic assays to minimize the number of false, negative results and improve the sensitivity of the diagnostic tests. Based on the results, it is recommended that the effects of TCFs on other mutants be investigated.

Keywords: COVID-19, Faradarmani, Taheri Consciousness Fields, T-Consciousness, Mutant, RT-PCR

INTRODUCTION

SARS-CoV-2 is a type of RNA virus. The process of virus replication is accompanied by a high mutation rate due to the lack of a mismatch repair mechanism. Hence, coronavirus mutations are quite predictable and reasonable. The virus may become more transmissible and difficult to eliminate due to mutations (Domingo et al., 1997). Korber et al. (2020) identified the D614G variant (the amino acid in the 614 position was mutated from aspartic acid to glycine), which was more contagious and had been dominated worldwide (Korber et al., 2020, Hou et al., 2020). Currently, other new strains are spreading rapidly worldwide, raising concerns about the prevention and treatment of COVID-19. Recent research has shown that only mutations with important biological functions show high transmissibility, suggesting that these key mutations may affect COVID-19 severity, virus spread, and escape from natural or vaccine-induced immunity (Zhou et al., 2021). SARS-CoV-2 infects human cells by binding to angiotensin-converting enzyme (ACE2) by the receptor-binding domain (RBD) of the Spike protein. These key mutations appear to affect the ability to bind to ACE2 (Starr et al., 2020).

Vaccination is carried out worldwide as a fundamental strategy to combat COVID-19. However, with the appearance of several variants of SARS-CoV-2, the effectiveness of vaccines has become a major topic of global debate. The current studies show that SARS-CoV-2 variants significantly affect the effectiveness of vaccines (Zhou et al., 2021).

Detection of COVID-19 is performed by SARS-CoV-2 RNA detection via real-time RT PCR. Studies on false-negative results from respiratory specimens for SARS-CoV-2 are varied and show a false-negative rate between 1 and 30% (Long et al., 2020, Arevalo-Rodriguez et al., 2020). The false-negative results can happen for several reasons, such as the non-optimal collection of samples, low analytical sensitivity, early testing of disease, unsuitable type of specimen, variability in the shedding of virus, or low viral load (Kinloch et al., 2020, Kucirka et al., 2020, Kanji et al., 2021, Pan et al., 2020). Given the urgency of controlling COVID-19 disease, it is necessary to find a novel way to control the mutation and infectivity of SARS-CoV-2 and decrease the false-negative cases of SARS-CoV-2 detection.

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 named Taheri Consciousness Fields (TCFs). In this perspective, T-Consciousness is one of the three existing elements of the universe apart from matter and energy. According to this theory, there are various TCFs with different functions, which are the subcategories of a networked universal internet called the Cosmic Consciousness Network (CCN). The major difference between the theory of TCFs and other theoretical concepts about consciousness is related to the practical application of the TCFs. TCFs can be applied to all living and non-living creatures, including plants, animals, microorganisms, materials, etc.

Mohammad Ali Taheri, the founder of Erfan Keyhani Halqeh, a school of thought, introduced a new science in 2020 as a branch of this school. He coined the term Sciencefact for this new science because it utilizes scientific investigations to prove the existence of T-Consciousness as an irrefutable phenomenon and a fact. Although science focuses solely on the study of matter and energy and Sciencefact, by contrast, explores the effects of the [non-material/non-energetic] TCFs, Sciencefact has provided a common ground between the two by conducting reproducible laboratory experiments in various scientific fields, and it has used the scientific approach in proving TCFs.

The influence of the TCFs begins with the Connection between CCN as the Whole Taheri Consciousness of the universe and the subjects of

study as a part. This Connection called "Ettesal" is established by a Faradarmangar's mind (a certified and trained individual who has been entrusted with the TCFs). The human mind has an intermediary role (Announcer) which plays a part by fleeting attention to the subject of study and then the main achievement obtained as a result of the effects of the TCFs. These fields cannot be directly measured by science, but it is possible to investigate their effects on various subjects through reproducible laboratory experiments (Taheri 2013).

The research methodology in the study of T-Consciousness has been founded on the process of *Assumption, Argument, and Proof*, in which the basic Assumption is: The Cosmos was formed by a third element called T-Consciousness that is different from matter and energy.

The Argument: The existence of TCFs can be demonstrated by its effects on matter and energy (e.g., humans, animals, plants, microorganisms, cells, materials, etc.)

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

Accordingly, to investigate and verify the existence, effects, and mechanisms of TCFs, the following five research phases (Phases 0 through 4), and the aims of each phase are outlined below.

Phase-0 studies aim to prove the existence of TCFs by observing their effects. The nature of T-Consciousness and what it is will not be addressed in this phase. Phase-1 explores the varied effects of different TCFs. Phase-2 examines the reason behind the varied effects of these fields. Phase-3 investigates the mechanism of TCFs effects on matter and energy. Finally, Phase-4 draws significant conclusions, particularly with regard to the mind and memory of matter and their relation to the T-Consciousness, etc.

In previous research, the effects of the TCFs on MCF7 cancer cell line (Taheri et al., 2020a), Alzheimer's disease rat models (Taheri et al., 2021b), spatial memory, and avoidance behavior of a rat model of Alzheimer's disease (Taheri et al., 2021c), wheat plant (Torabi et al., 2020), bacterial population growth (Taheri et al., 2021d), viral growth (Taheri et al., 2021a), and the electrical activity of the brain during the Faradarmani Connection in the Faradarmangars population (Taheri et al., 2020b) have been investigated.

The present study aimed to evaluate the effect of Faradarmani CF as one of the TCFs on mutant D614G virus titer and RNA copy number and the quality of amplification of coronavirus genome fragments.

MATERIALS AND METHODS

Application of Faradarmani Consciousness Fields

Subjects of the study were influenced by TCFs according to the protocols mentioned on the website of research management in the TCFs (www.COSMOintel.com). Requesting an announcement is free of cost (can be found in the "Assign Announcement" section). The researchers introduce the test to the COSMOintel center after registration on the mentioned website to conduct studies at any time and place. For example, the number of samples, controls and their contractual name must be specified. It should be mentioned that this study was conducted in a double-blinded way. It means that the experts were completely unfamiliar with TCFs theory. Also, the person who established the Connection was unfamiliar with the details of this study. In the present study, Faradarmani CF was announced exactly at the same time as the virus inoculation in cell culture flasks in treatment groups of examinations.

Preparation of mutant and non-mutant strains

For the current study, non-mutant strains similar to the original Wuhan strain, as well as mutant strains with D614G mutation in virus spike pro-

tein, were used. The above strains were isolated from the nasopharyngeal and oropharyngeal hospital specimens of patients admitted to the intensive care unit in the acute phase of the disease with positive real-time RT-PCR results and Ct values of 13 and 12.

Cell culture and virus titration

The T-25 flasks were seeded with 5×10^6 Vero cells in culture media composed of high glucose DMEM (Gibco) with 10% fetal bovine serum (Gibco) and incubated in 5% CO_2 at 37°C until 80% confluency. Then they were divided into five groups with six flasks to test the mutant virus and non-mutant virus with and without the Faradarmani CF treatment as well as negative control (without virus). Cell cultures were prepared for virus inoculation after reaching 70% confluency. A biosafety level 3 (BSL-3) laboratory was used for all surveys on the virus (WHO 2020). The virus culture with TCID50/ml~log 6 was selected for inoculations into the flasks. TCID50/ml was assessed in 96 well plates, and all the plates were monitored every 24 h for CPE (Cytopathic effect), and after four days, results were reported, and the virus titer calculations were performed using Read & Munch method (Reed et al., 1938).

Real-time RT-PCR for evaluation of SARS-CoV-2 virus RNA copy number

LabPrep™ Viral DNA/RNA Mini Kit was used for extraction of RNA according to the manufacturer's protocol. SinaClone cDNA synthesis kit was used for cDNA synthesis. All steps were performed with RNase-free tools and solutions. Also, in order to make cDNAs with the same concentration in each sample, according to the concentration of RNA, the volume was taken in which the amount of 1000 ng of RNA was present, and with water and Master Mix, the final volume was reached 20 microliters. Biotechrabbit GmbH kit was used for real-time PCR testing. RNA copy numbers were evaluated using Nucleocapsid (N) gene primers according to the protocol and material. Rotor-Gene-Q 6000 thermocycler (Corbett, Australia) was used for performing all real-time PCR reactions.

RT-PCR and electrophoresis of PCR products of S protein fragments of mutated SARS-CoV-2

For six selected and positive real-time PCR samples with a Ct value of 13 for which RNA was extracted and cDNA was previously fabricated, and with three pairs of primers called B26, B27, and B28 for different parts of S1 spike protein, PCR was performed (Table 1).

PCR products were run on 1% agarose electrophoresis gel in TBE buffer containing Tris base, boric acid, EDTA, and ethidium bromide with pH=8. For electrophoresis, the products were exposed to a voltage of 90 volts for 55 minutes, using a 100 bp Plus DNA Ladder (Germany, Fermentas) as a marker.

RESULTS

Faradarmani CF increased replication of non-mutated and mutated viruses compared with control samples. Moreover, The Faradarmani CF increased the replication rate of the mutant strain close to the non-mutant strain in the cells (Table 2). There was an increase of 0.7 logs, which is about five times in replication of the non-mutated virus. Faradarmani CF increased the mutated virus by 0.2 logs (log 7 to 7.2 log) which increased replication by 1-fold (3 to 2), which slightly in-

Table1. Sequences of applied primers for PCR of S1 spike protein.

Primer number	Sequence of primer	Length	Annealing temp (°C)	Target protein	Ref
B26	F: TATCTTGGCAAACCACGCGA R: ACCAGCTGTCCAACCTGAAG	1057	58	Spike	(Ren et al., 2020)
B27	F: CCCTCAGGGTTTTTCGGCTT R: CTGTGGATCACGGACAGCAT	1093	60	Spike	(Ren et al., 2020)
B28	F: CCAGCAACTGTTTGTGGACC R:GTGGCAAAACAGTAAGGCCG	1027	60	Spike	(Ren et al., 2020)

creased the mutated virus titer. Also, Faradarmani CF increased the time required for cell destruction and mortality.

In real-time PCR results, the RNA copy number of the non-mutant control group with the non-mutant group affected by Faradarmani CF was decreased with a difference of 3 Ct value. Also, in the mutant group, this difference between the control mutant and mutant affected with Faradarmani CF was decreased with a difference of one Ct value.

To investigate the effect of the Faradarmani CF on the RT-PCR of mutated coronavirus genome and the amplification rate of coronavirus genome fragments, PCR was performed on three important fragments of the coronavirus glycoprotein spike gene responsible for cell binding. Then gel electrophoresis was done on PCR products. The sharper detected bands in the samples treated with Faradarmani (data not shown) might signify the effect of Faradarmani on the quality of amplification in the PCR, but for proving this assumption needs to do complementary experiments.

DISCUSSION

The D614G substitution on the spike glycoprotein in the Wuhan primary strain increased the virus replication (Plante et al., 2021). Plante et al. (2021) displayed the importance of the G614 variant in spreading the virus and its implications for the efficacy of vaccines and therapy with antibodies. The G614 virus replicated more than the original D614 virus in primary human upper airway tissues and the human airway epithelial cell line (Calu-3). The increased replication fitness was associated with increases in specific stability and infectivity of the G614 virus (Plante et al., 2021). The higher levels of viral RNA were detected in patients infected with the D614G mutant, and this mutant is more infective than the original Wuhan strain (Korber et al., 2020). Ozono et al. (2021) showed that the D614G mutation confers increased entry efficiency resulting from the enhanced binding affinity for ACE2 with no influence on the antigenicity of the S protein (Ozono et al., 2021).

This study showed that the Faradarmani CF increased replication of non-mutated and mutated viruses compared to the group without Faradarmani CF treatment and increased replication fitness, and the virus gained more control over the cellular replication machinery. It seems that the Faradarmani CF increases the rate of replication of the mutant strain close to the non-mutant strain in the cells and reaches a biological equilibrium of compatibility similar to that of the Wuhan strain with the cell. In order to elucidate the effects of this treatment, we recommend that the influence of the TCFs on in-vivo animal models with mutated and non-mutated virus infection be evaluated.

False-negative results have vital implications for the isolation and transmission risk and for the management of COVID-19 (West et al., 2020). According to the results of the Faradarmani CF effect on the RT-PCR of mutated coronavirus genome, Faradarmani CF may improve different steps of PCR such as primer binding, amplification, and electrophoresis motion. According to the results, the Faradarmani CF may have an effective role in the PCR processes of virus fragments and can be helpful in the increase of laboratory tests accuracy and decrease false-negative results.

As it was mentioned, TCFs are not measurable, but it is possible to investigate their effects indirectly through various experiments. We recommend the investigation of other mutations of the COVID-19 virus to study the effect of the Faradarmani CF on the success of immunization and vaccination in vivo.

Table2. The titer of mutant and non-mutant SARS-CoV-2 under influence of Faradarmani CF and control groups. All conditions, except virus strains, were the same for both Faradarmani CF groups.

	Inoculum (Seed)	Non-mutated virus	Mutated virus (control)	Non-mutated virus+ FCF	Mutated Virus+ FCF	Cell culture (Cell control)
TCID50/mL	1×10^{6}	$2\times10^{6.5}$	3×10^{7}	$2\times10^{7.2}$	$2\times10^{7.2}$	-
Ct Value in real-time RT-PCR	14	11	9	8	8	-

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CONFLICTS OF INTEREST

The authors declare no conflict of interest.

REFERENCES

Arevalo-Rodriguez I., Buitrago-Garcia D., Simancas-Racines D., Zambrano-Achig P., Del Campo R., Ciapponi A., Sued O., Martinez-Garcia L., Rutjes A. W. & Low N. (2020). False-negative Results of Initial RT-PCR Assays for COVID-19: A Systematic Review. *PLoS One*, 15, e0242958.

Domingo E. & Holland J. (1997). RNA Virus Mutations and Fitness for Survival. *Annual Review of Microbiology*, 51, 151-178.

Hou Y. J., Chiba S., Halfmann P., Ehre C., Kuroda M., Dinnon K. H., Leist S. R., Schäfer A., Nakajima N. & Takahashi K. (2020). SARS-CoV-2 D614G Variant Exhibits Efficient Replication Ex vivo and Transmission in Vivo. *Science*, 370, 1464-1468.

Kanji J. N., Zelyas N., MacDonald C., Pabbaraju K., Khan M. N., Prasad A., Hu J., Diggle M., Berenger B. M. & Tipples G. (2021). False Negative Rate of COVID-19 PCR Testing: A Discordant Testing Analysis. *Virology Journal*, 18, 1-6.

Kinloch N. N., Ritchie G., Brumme C. J., Dong W., Dong W., Lawson T., Jones R. B., Montaner J. S., Leung V. & Romney M. G. (2020). Suboptimal Biological Sampling as a Probable Cause of False-negative COVID-19 Diagnostic Test Results. *The Journal of Infectious Diseases*, 222, 899-902.

Korber B., Fischer W. M., Gnanakaran S., Yoon H., Theiler J., Abfalterer W., Hengartner N., Giorgi E. E., Bhattacharya T. & Foley B. (2020a). Tracking Changes in SARS-CoV-2 Spike: Evidence that D614G Increases Infectivity of the COVID-19 Virus. *Cell*, 182, 812-827. e19.

Kucirka L. M., Lauer S. A., Laeyendecker O., Boon D. & Lessler J. (2020). Variation in False-negative Rate of Reverse Transcriptase Polymerase Chain Reaction–based SARS-CoV-2 Tests by Time Since Exposure. *Annals of Internal Medicine*, 173, 262-267.

Long D. R., Gombar S., Hogan C. A., Greninger A. L., Shah V. O. R., Bryson-Cahn C., Stevens B., Rustagi A., Jerome K. R. & Kong C. S. (2020). Occurrence and Timing of Subsequent SARS-CoV-2 RT-PCR Positivity Among Initially Negative Patients. *Clinical Infectious Diseases*.

Ozono S., Zhang Y., Ode H., Sano K., Tan T. S., Imai K., Miyoshi K., Kishigami S., Ueno T. & Iwatani Y. J. N. c. (2021). SARS-CoV-2 D614G Spike Mutation Increases Entry Efficiency with Enhanced ACE2-binding Affinity. 12, 1-9.

Ozono, S., Zhang, Y., Ode, H., Sano, K., Tan, T. S., Imai, K., Miyoshi, K., Kishigami, S., Ueno, T., Iwatani, Y., Suzuki, T., & Tokunaga, K. (2021). SARS-CoV-2 D614G Spike Mutation Increases Entry Efficiency with Enhanced ACE2-binding Affinity. *Nature Communications*, 12(1), 848.

Pan Y., Long L., Zhang D., Yuan T., Cui S., Yang P., Wang Q. & Ren S. (2020). Potential False-negative Nucleic Acid Testing Results for Severe Acute Respiratory Syndrome Coronavirus 2 from Thermal Inactivation of Samples with Low Viral Loads. *Clinical Chemistry*, 66, 794-801.

Plante J. A., Liu Y., Liu J., Xia H., Johnson B. A., Lokugamage K. G., Zhang X., Muruato A. E., Zou J. & Fontes-Garfias C. R. J. N. (2021). Spike Mutation D614G Alters SARS-CoV-2 Fitness. 592, 116-121.

Reed L. J. & Muench H. (1938). A Simple Method of Estimating Fifty Percent Endpoints. *American Journal of Epidemiology*, 27, 493-497.

Ren L.-L., Wang Y.-M., Wu Z.-Q., Xiang Z.-C., Guo L., Xu T., Jiang Y.-Z., Xiong Y., Li Y.-J. & Li X.-W. (2020). Identification of a Novel Coronavirus Causing Severe Pneumonia in Human: A Descriptive Study. *Chinese Medical Journal*, 133, 1015.

Starr T. N., Greaney A. J., Hilton S. K., Ellis D., Crawford K. H., Dingens A. S., Navarro M. J., Bowen J. E., Tortorici M. A. & Walls A. C. (2020). Deep Mutational Scanning of SARS-CoV-2 Receptor Binding Domain Reveals Constraints on Folding and ACE2 Binding. *Cell*, 182, 1295-1310. e20.

Taheri M. A. (2013). *Human from Another Outlook* (2nd Edition). *ISBN-13: 978-1939507006, ISBN- 10: 1939507006*.

Taheri M. A., Etemadi M. R., Torabi S., Nabavi N. & Semsarha F. (2021a). Evaluation of the Influence of Faradarmani Consciousness Field on Viral Growth.

Taheri M. A., Semsarha F., Mahdavi M., Afsartala Z. & Amani L. (2020a). The Influence of the Faradarmani Consciousness Field on the Survival and Death of MCF-7 Breast Cancer Cells: An Optimization Perspective. *Available at SSRN 3705537*.

Taheri M. A., Semsarha F. & Modarresi-Asem F. (2020b). An Investigation on the Electrical Activity of the Brain during Faradarmani Connection in the Fara-Therapist Population.

Taheri M. A., Torabi S., Nabavi N. & Semsarha F. (2021b). Faradarmani Consciousness Field Suppresses Alzheimer's Disease Development in Both in Vitro and in Vivo Models of The Disease.

Taheri M. A., Torabi S., Nabavi N. & Semsarha F. (2021c). Influence of Faradarmani Consciousness Field on Spatial Memory and Passive Avoidance Behavior of Scopolamine Model of Alzheimer Disease in Male Wistar Rats.

Taheri M. A., Zarrini G., Torabi S., Nabavi N. & Semsarha F. (2021d). Influence of Faradarmani Consciousness Field on Bacterial Population Growth. *BioRxiv*.

Torabi S., Taheri M. A. & Semsarha F. (2020). Alleviative Effects of Faradarmani Consciousness Field on Triticum Aestivum L. Under Salinity Stress. *F1000Research*, 9, 1089.

West C. P., Montori V. M. & Sampathkumar P. (2020). COVID-19 Testing: The Threat of False-negative Results. In *Mayo Clinic Proceedings*, 1127-1129. Elsevier.

World Health Organization. (2020). Laboratory Biosafety Guidance Related to Coronavirus Disease 2019 (COVID-19): Interim Guidance, 12 February 2020. World Health Organization. https://apps.who.int/iris/handle/10665/331138.

Zhou W. & Wang W. (2021). Fast-spreading SARS-CoV-2 Variants: Challenges to and New Design Strategies of COVID-19 Vaccines. *Signal Transduction and Targeted Therapy*, 6, 1-6.