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Evaluation of the Effect of Taheri Consciousness Field 1 on Nanostructured Lipid Carriers in the Treatment of UV-irradiated Human Hair Keratin

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Abstract

Nanostructured lipid carriers (NLCs) are self-assembled structures formed from nanoemulsions of mixed solid and liquid lipids and are used in drug delivery systems. Research on NLCs has shown that these carriers offer advantages over conventional carriers for drug therapy, including increased solubility, stability, improved permeability, and enhanced bioavailability of drugs. The process of self-organization has attracted considerable research interest due to its potential role in the origin of life. Furthermore, it has been shown that solar ultraviolet (UV) waves play two contradictory roles in the prebiotic and biological atmosphere during the process of life emergence: a constructive role in providing the initial energy for reactions effective in forming organic molecules, and a destructive role in breaking down biological molecules in the early ozone-free atmosphere, which was rich in disruptive radiation. Considering the importance of self-assembled structures in both the origin of life and drug delivery, as well as the significant effects of T-Consciousness Field 1 (TCF1) on the structure and function of enzymes, biomimetic molecules, and biological cells observed in previous studies, the present research was conducted to investigate the effect of TCF1 on human hair keratin exposed to ultraviolet radiation. Based on the obtained results, the diameter of the NLCs and the zeta potential of the sample treated with TCF1 increased by approximately 7% and 100%, respectively, compared to the control. The PDI (polydispersity index) of the sample also decreased by about 31% compared to the control. Moreover, the use of NLCs synthesized under treatment with TCF1 led to significant repair and improvement of keratin protein exposed to UV radiation. Further studies are needed to provide deeper insight into the potential effects of TCF1 on biological and self-assembly systems.

Keywords: Nanostructured lipid carriers, Ultraviolet radiation, Self-organization, T-Consciousness Field, Origin of life, Keratin

Introduction

Nowadays, the use of nanotechnology provides the opportunity to enhance treatments through targeted drug delivery. For example, this application can increase drug flux into and through the skin (Goyal et al., 2016). Nanoparticles are defined as tiny particles with dimensions between 1 and 100 nm (Khan et al., 2019). Solid lipid nanoparticles (SLNs) were conceptualized by Müller and Gasco as a potential delivery particle system in the early 1990s. These nanocarriers (50–1000 nm) offer a great number of advantages; for instance, they have been utilized in various drug deliveries (Yetisgin et al., 2020) and as biocompatible lipids (Lacatusu et al., 2011). Furthermore, minimal use of organic solvents in the formulation and high in vivo stability are other benefits of using SLNs (Uner et al., 2007). However, they have certain limitations, such as poor drug-loading capacity, unpredictable gelling tendency, and drug leakage (Mishra et al., 2018). Nanostructured lipid carriers (NLCs) have also been introduced after SLNs to overcome their possible shortcomings. NLCs are composed of physiological and biocompatible lipids, surfactants, and co-surfactants (Chauhan et al., 2020). They provide better stability and loading capacity, and they prevent drug expulsion during storage (Naseri et al., 2015).

The main characteristic feature of nanoparticles in a colloidal medium, which indicates their stability in an aqueous medium for a biomedical application, is the zeta potential (ζ [=] mV) of the particles (Gordillo-Galeano and Mora-Huertas, 2021). The zeta potential is often considered the effective charge on the particle (Hiemenz & Rajagopalan, 1997). Particles of the same charge with increasing absolute zeta potential will have less tendency to aggregate, and generally, suspensions with a zeta potential of $\zeta > |30|$ are considered suspensions with good colloidal stability (Freitas & Müller, 1998).

Self-organization is a phenomenon in which components of a system assemble through noncovalent forces such as in micelles (Förster et al., 2002). Not only does self-organization play a crucial role in building up complex structures, but it also provides a fundamental process in the emergence of life (Mann, 2012; Li, 2017; Mendes et al., 2013). The origin of life is one of the most important challenges in science. There have been many efforts to solve this problem for nearly 100 years (Oró et al., 1990). In most theories and experiments in this context, ultraviolet waves have played a key role in the formation of life (Miller & Urey, 1959; Oparin, 1965; Lazcano, 2010).

Undeniably, water molecules are considered an essential factor in the origins of life. They have been described by their ambivalent role. This means that, in addition to acting as a solvent and reactant, they also enhance hydrolysis, which is a powerful force in promoting self-organization in prebiotic chemistry (do Nascimento Vieira et al., 2020). Considering the destructive role of UV in breaking the primary bonds of organic molecules in early life, water has been suggested as a possible candidate to protect against these effects (Ranjan et al., 2016). It should be noted that, in addition to the biochemical study of life, we need to understand the physical processes and driving forces to know how the transition of matter occurred from the nonliving to the living state (Cronin and Walker, 2016; Goldenfeld and Woese, 2011). Life cannot be explained by our current laws of physics and requires new physics to probe the origins of life (Walker, 2019).

According to Taheri's theory, there are diverse T-Consciousness Fields (TCFs) with various functions that are subsets of the Cosmic Consciousness network. These fields are non-frequency in nature and cannot be measured with quantitative tools, but it is possible to record their effects in the laboratory. In general, from this perspective, T-Consciousness is considered a fundamental component of the universe, from which matter, energy, and information originate (Taheri, 2013). Therefore, this theory suggests that consciousness played a key role at the beginning of the emergence of life.

The use of TCFs allows us to investigate the potential interactions of consciousness with biomimetic models. These studies can serve as a foundation for future research in this area. Previously, the

effects of TCF1 on the structure and function of horseradish peroxidase, as well as gold nanozyme and supramolecular micellar biomimetic models, have been examined, and the results were published in this same issue. Moreover, several studies have reported the effects of TCFs on various subjects, including plant and animal models (Torabi et al., 2020; Taheri et al., 2021).

Keratin, as a structural protein, is rich in the amino acid cysteine, which is covalently linked via disulfide bonds with crosslinks between strands, making it one of the strongest non-mineralized tissues in nature (Zhang et al., 2013). In the present study, self-organized structured NLCs were utilized with two objectives: first, to investigate the role of TCF1 in repairing damage caused by ultraviolet radiation in the Earth's early atmosphere, and second, to explore its possible role in the optimal organization of early life forms. Additionally, the alleviative effect of TCF1 on UV-irradiated hair keratin was investigated.

Materials and Methods

Application of TCF1

TCF1 was applied to the samples according to the protocol established by Cosmointel Research Center[4]. Further details are provided in the general consideration of this issue.

NLCs were synthesized by emulsion and ultrasound homogenization. First, the oil phase (consisting of solid and liquid lipids) was heated in a water bath to 5 °C above the lipid melting point (75 °C). Then, the aqueous phase (containing the surfactant Tween 80 and deionized water) at 75 °C was added dropwise to the lipid phase using a syringe pump, under magnetic stirring at 1200 rpm. At this stage, a coarse oil-in-water emulsion, consisting of large droplets of solid and liquid lipids, was formed and then further refined into fine droplets using a magnetic stirrer (Fan Azma Gostar) at 1200 rpm. Finally, to reduce nanoparticle size, the synthesized formulation was transferred to a homogenizer (developed using Iranian ultrasonic technology) and homogenized for 15 min with cycles of 10 second of homogenization followed by 3 second of rest. The emulsion was then placed in an ice bath for 10 minutes to induce the formation of incomplete lipid nanostructured crystals. Due to the formation of a surfactant layer around each lipid nanostructure, creating a spatial barrier between them, stable dispersed nanostructured lipid carriers were obtained in the aqueous medium after cooling and crystal formation.

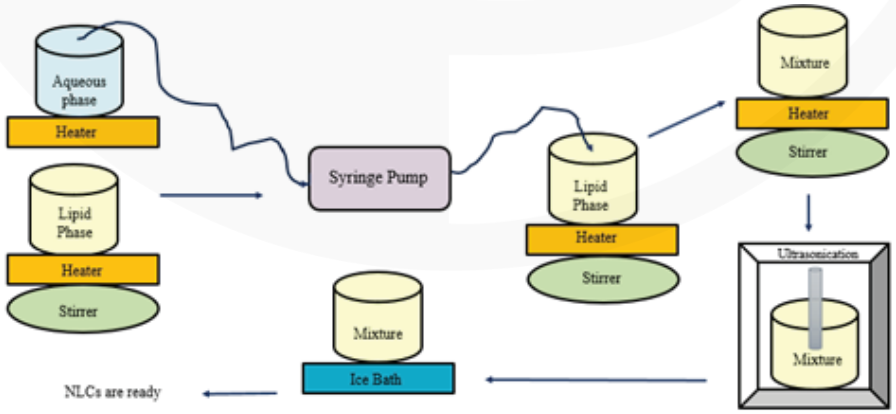


Figure 1. Synthesis process of NLC in the present study.

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Dynamic Light Scattering

The mean particle size, polydispersity index (PDI), and surface charge of the lipid nanostructures were measured using a dynamic light scattering device (Brookhaven/Plus Pals-90, USA). All samples were diluted with deionized water. Particle size and the scattering index were determined using the device in automatic measurement mode at a fixed angle of 90° and a temperature of 25 °C, while the surface charge (zeta potential) was calculated based on electrophoretic mobility.

Preparation and Treatment of Hair Samples

Before performing functional assays, all hair samples were washed with a non-ionic detergent (baby hair shampoo) and water (Shavandi et al., 2017). A 0.05 g portion of black natural human hair fiber was used as a control (intact and not subjected to TCF1), and the same amount was used as a sample (damaged by ultraviolet light and treated with NLCs prepared under the influence of TCF1). The procedures for inducing damage in hair keratin, protein extraction, and concentration assays are described as follows.

UV Irradiation of Human Hair

After treating the hair with the synthesized NLC solution under the influence of TCF1, the samples were irradiated for six hours using a modeled source of UVA and UVB (Osram) ultraviolet radiation. Following irradiation, the hair was washed again with a non-ionic shampoo (baby hair shampoo) and further dried before analysis (Prasertpol & Tiyaboonchai, 2020). Finally, external lipids and hair fiber contaminants were removed by immersing the hair in distilled water for one hour, followed by treatment with a 2:1 v/v chloroform: methanol (merc) solution for 24 hours (Shavandi et al., 2017).

Protein Extraction and Assay

Preparation of Buffer Solution

Tris-HCl (25 mM), thiourea (2.6 M), urea (5 M), and 2-mercaptoethanol (Merc) were used to prepare the buffer solution. To prepare 160 ml of buffer, 1.21 g of Tris powder was dissolved in 20 ml of deionized water, followed by the addition of 200 µl of HCl to adjust the solution to pH 8.5. The volume of this Tris-HCl solution was then brought to 40 ml. Separately, 7.9 g of thiourea was dissolved in 40 ml of deionized water, and 12 g of urea was added, bringing the volume of the final solution to 80 ml. A 5% mercaptoethanol solution was prepared by mixing 2 ml of mercaptoethanol with 38 ml of deionized water, and this was added to the solution containing Tris-HCl, thiourea, and urea. Finally, the resulting buffer solution was homogenized using a Vertex mixer (Shavandi et al., 2017; Nakamura et al., 2002; Cassoni et al., 2018).

Extraction of Keratin

Hair samples previously treated with chloroform/ methanol solution were washed with distilled water and then dried. The samples were cut into pieces 1–2 mm in length and immersed in 5 ml of the prepared buffer solution (described in the previous section). They were then incubated in a Binder incubator (Germany) at 48 °C for 48 hours. In the final step, the solution was filtered using a 0.2 µm filter (Sartorius, Germany) and centrifuged (Hitachi, Japan) at 16,000 × g for 15 min at room temperature. The resulting supernatant was collected and used as hair protein (keratin) (Cassoni et al., 2018).

Determination of Keratin Concentration by Bradford Protocol

The ingredients for this assay included a standard BSA solution (Sigma) with a concentration of 1 mg/ml, from which solutions with concentrations of 0.02, 0.04, 0.06, 0.08, and 0.1 mg/ml were prepared. The Bradford reagent was prepared as follows: first, 10 mg of Coomassie Brilliant Blue G-250 was dissolved in 10 ml of methanol; second, 10 ml of 85% H_3PO_4 and 50 ml of deionized water were added to the solution; third, the solution was filtered; and finally, the volume was brought to 100 ml. The Bradford solution was stored at 4 °C in the dark. The assay was performed as follows: 100 µl of each standard and unknown sample solution was pipetted into microplate wells, followed by 100 µl of Bradford reagent. The samples were gently mixed by shaking the microplate and incubated at room temperature for at least five minutes (Bradford,

1976). The absorption intensity of the blue dye was used to determine the protein content in the solution with a microplate reader (PowerWave XS2, BioTek, USA) at 595 nm. The concentration of keratin was calculated using a standard BSA curve in the concentration range of 20–100 µg/ml (Bradford, 1976).

Statistical Analysis

In this study, each sample had three replicates, and no data were excluded. Comparisons between the test and control samples were analyzed using the t-test in SPSS software, version 17, and a confidence level of 95% was considered statistically significant.

Results

Structural Analysis of the NLC Solution

The results of elastic light scattering for the sample and control in this study are presented in Table 1. The particle size distribution data for the sample and control, obtained using the DLS method, are shown in Figures 2 and 3. As indicated in Table 1, the diameter of the NLCs and the zeta potential of the sample increased by approximately 7% and 100%, respectively, compared to the control. Additionally, the PDI (polydispersity index) of the sample decreased by about 31% compared to the control.

Table 1. Data obtained from the measurement of colloidal properties of the sample treated with TCF1 (Sample) and the control using the DLS technique.

Name	Particle Size range/nm	Effective diameter/nm	Mean Diameter/ nm	Var.	Max Size/ nm	PDI (Polydispersity Index)	Zeta potential/ mV	Mobility
Control	74.01-106.83	350.0	84.1	0.1	81.13	0.32±0.01	-16.23±1.49	-1.27
Sample	70.07-106.11	432.9	89.69	0.5	80.46	0.22±0.02	-33.10±0.49	-2.59

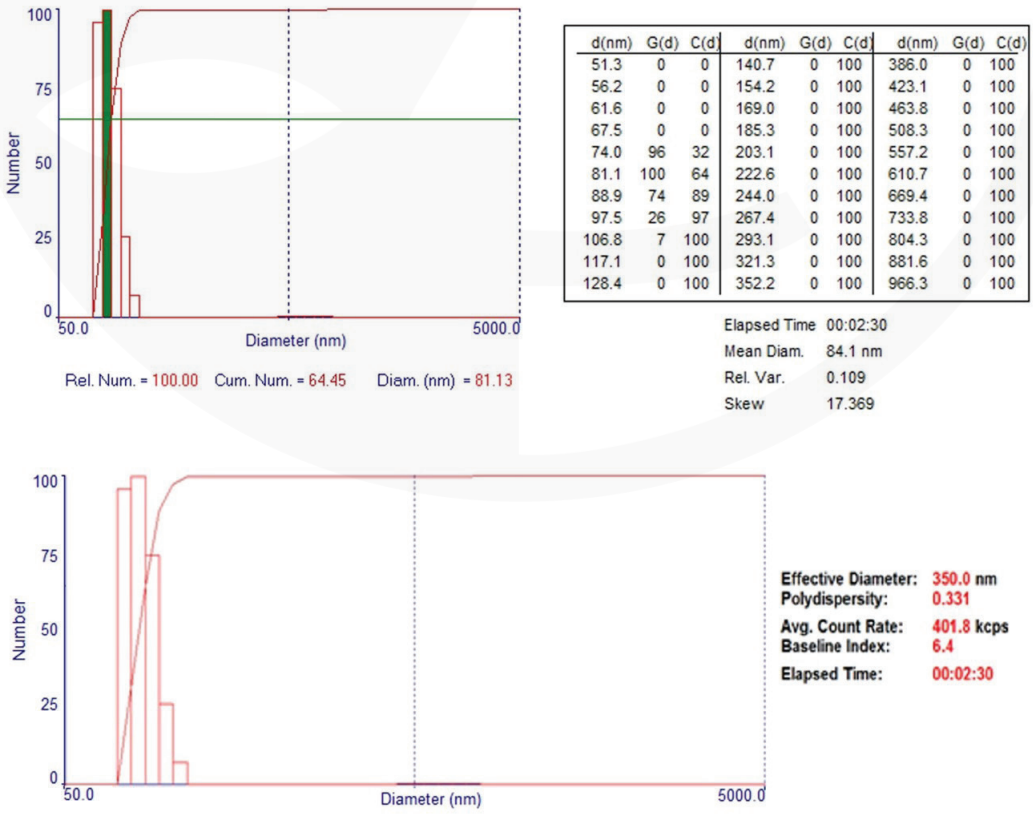
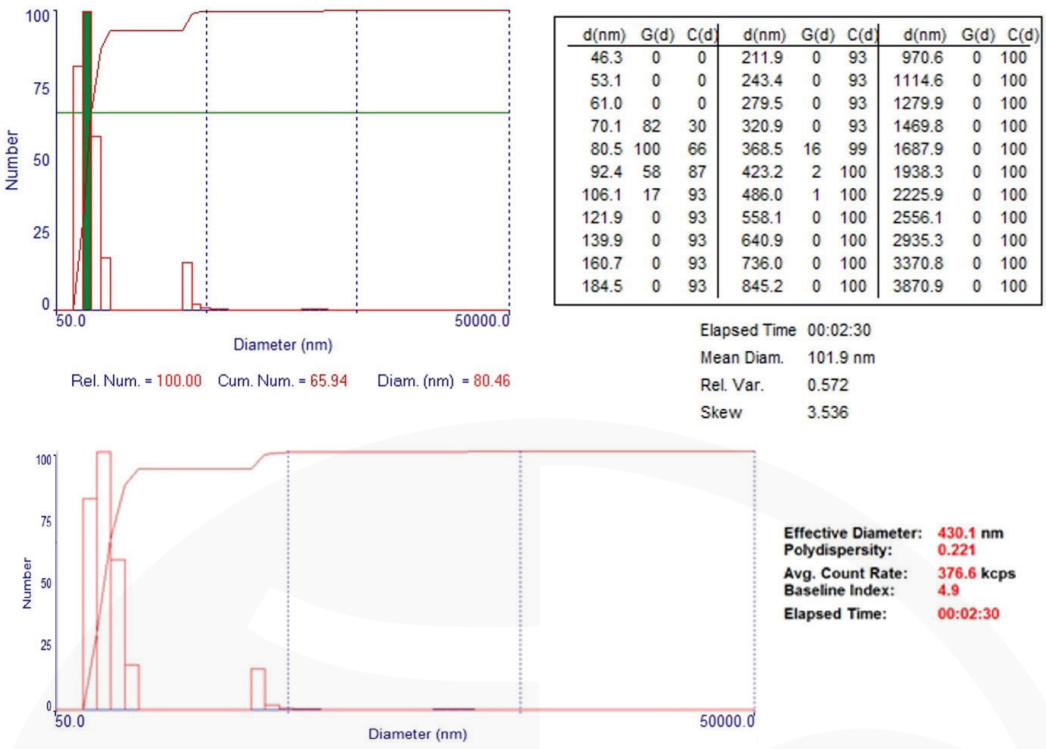


Figure 2. Particle size distribution of NLCs in the control.



d(nm)	G(d)	C(d)	d(nm)	G(d)	C(d)	d(nm)	G(d)	C(d)
46.3	0	0	211.9	0	93	970.6	0	100
53.1	0	0	243.4	0	93	1114.6	0	100
61.0	0	0	279.5	0	93	1279.9	0	100
70.1	82	30	320.9	0	93	1469.8	0	100
80.5	100	66	368.5	16	99	1687.9	0	100
92.4	58	87	423.2	2	100	1938.3	0	100
106.1	17	93	486.0	1	100	2225.9	0	100
121.9	0	93	558.1	0	100	2556.1	0	100
139.9	0	93	640.9	0	100	2935.3	0	100
160.7	0	93	736.0	0	100	3370.8	0	100
184.5	0	93	845.2	0	100	3870.9	0	100

Figure 3. Particle size distribution of NLCs in the sample treated with TCF1.

Functional Analysis of NLC Particles

The effects of TCF1 on hair exposed to ultraviolet radiation (via NLC particles treated with TCF1 during synthesis) were evaluated by measuring keratin protein absorption at a wavelength of 595 nm. The absorbance and keratin concentration in the sample and control are presented in Figure 4 and Table 2, respectively.

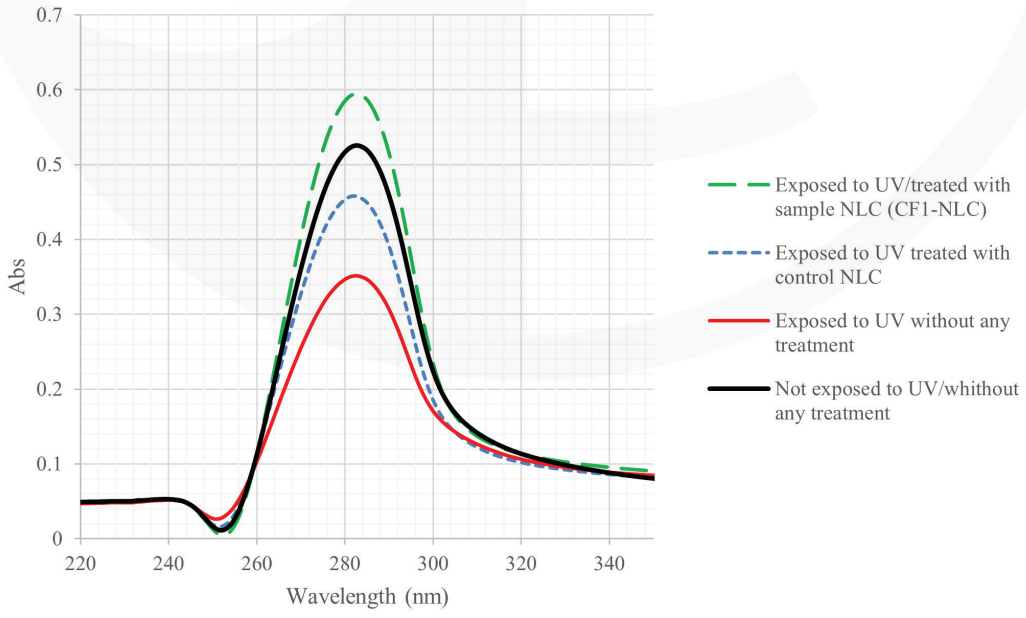


Figure 4. Keratin spectrophotometry in the sample treated with TCF1-NLC (CF1-NLC) and the control.

Table 2. λmax and calculated keratin concentration in the sample and control (obtained from the data in Figure 4).

Hair sample	$\lambda_{max/nm}$	Intensity	Keratin Concentration (µg/ml)	Change in comparison with natural hair (%)
Exposed to UV radiation-treated with sample NLC	283	0.594	69.7±0.2	**+16**
Not exposed to UV/without NLC treatment (natural healthy hair)	283	0.525	59.9±0.1	-
Exposed to UV radiation-treated with control NLC	282	0.457	50.8±0.3	**-15**
Exposed to UV/without NLC treatment	282	0.351	38.0±0.2	**-40**

As shown in Figure 4 and Table 2, hair samples exposed to ultraviolet radiation exhibited hyperchromism and a blue shift under the influence of NLC particles treated with TCF1. This indicates that, in addition to repairing UV-induced damage, the treatment resulted in an improved structure and an increased keratin protein concentration, even compared to the intact control sample.

Discussion

The application of NLCs as self-assembling drug carrier nanoparticles has received considerable attention. Since they are non-toxic or exhibit very low toxicity, they can prolong the drug's action through an extended half-life and controlled release (García-Pinel et al., 2019). These compounds are formed from surfactants and lipids in different phases within an aqueous environment (Bhaskar et al., 2009). Water molecules at the core of these structures can transport various hydrophilic drug compounds. This study utilized the self-assembling properties of these compounds and the important role of water in their formation and structure.

In the first stage of this study, the effect of TCF1 on the structure and aggregation of NLC particles in a colloidal medium was investigated using the DLS technique. The polydispersity index (PDI) is an important parameter that describes the particle size distribution and ranges from 0 to 1. A PDI below 0.1 indicates a monodisperse particle population, while values above 0.1 indicate a polydisperse size distribution. In addition to particle size, the zeta potential (ZP) is used as an indicator of an effective and stable nanoparticle system (L. Kiss et al., 2019). This parameter reflects the repulsion between charged particles in the dispersion. A high ZP indicates more highly charged particles, which prevents particle aggregation due to electrostatic repulsion. Conversely, if the ZP is low, van der Waals forces dominate over electrostatic repulsion, increasing the likelihood of mixture coagulation. A zeta potential value of ±30 mV is considered indicative of good particle distribution stability in nanoparticles (Pate and Safier, 2016).

Based on the results of this study, the polydispersity of the sample treated with TCF1 decreased by approximately 31% compared to the control, indicating a distribution closer to monodispersity. In addition, the zeta potential of the sample increased to more than twice that of the control, reflecting significant stability and approaching the optimal zeta potential value for nanoparticles. Interestingly, no significant change in particle size was observed in the sample compared to the control (approximately 6%). This result suggests that TCF1 altered system properties such as surface charge without affecting the overall dimensions of each particle.

In the second stage, it was observed that these structural changes in NLCs synthesized under the influence of TCF1 had a significant impact on their performance in treating keratin in hair exposed to UV radiation. The results showed that TCF1 not only repaired the damaged hair samples but also optimized the structure of the target protein, making it 16% healthier than the control not exposed to ultraviolet radiation.

According to the theory of T-Consciousness Fields, the influence of these non-physical fields occurs through the transmission of information from them to the subject under study, resulting in observable changes in the behavior of the subject. These results provide evidence of such effects. In this study, the overall aggregation of NLC particles and the water present at their core were effectively used as carriers of information transmitted from TCF1 to repair the system under investigation. As mentioned in the introduction, in this view, T-Consciousness is a fundamental component of the universe and the source of matter, energy, and information. Therefore, it has played a key role in the formation and evolution of life. Considering the self-assembling properties of NLCs and their significant resemblance to primitive membrane structures, which were exposed to destructive ultraviolet waves at the origin of life, this study preliminarily examined the enhancing effects of TCF1 and, using this model, explored the hypothesis regarding the role of consciousness and these non-frequency fields under early-life conditions.

In summary, this study provides evidence of the enhancing effect of TCF1 on UV-irradiated hair keratin protein and the resulting changes in the properties of particles influenced by this field. From Taheri's perspective, these changes are attributed to the transfer of information via T-Consciousness Fields. To gain a better understanding of the mechanisms and effects of these fields, as well as to explore their potential role in the origin of life, further studies are planned by the authors.

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