Effect of The Biofield Energy Healing Treatment on the Pharmacokinetics of 25-Hydroxyvitamin D$_3$ [25(OH)D$_3$] in Rats After a Single Oral Dose of Vitamin D$_3$

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Abstract: There are numerous scientific studies on vitamin D$_3$ reported to have many important roles viz. in bone metabolism, osteoporosis, immunity, and useful in several diseases, e.g., cancers, cardio-vascular, and neurodegenerative diseases. Surprisingly, there is little information available about the concentration of 25-hydroxyvitamin D$_3$ in the blood, which is the best indicator of vitamin D$_3$ status after its oral absorption. However, the biological activity of vit-D$_3$ is mediated via the formation of the active metabolite, 1-α, 25-dihydroxyvitamin D$_3$. There are several factors which affect its absorption and first-pass metabolism that lead to have very low plasma concentrations of both the metabolites (25-hydroxyvitamin D$_3$ and 1-α, 25-dihydroxyvitamin D$_3$) following an oral administration. Therefore, the present study was performed to determine the effects of The Trivedi Effect®-Energy of Consciousness Healing Treatment on vit-D$_3$ and rats through the measurement of plasma 25-hydroxyvitamin D$_3$ concentrations after the oral administration of vit-D$_3$ in rats. The test item, vit-D$_3$ was divided into two parts. One part was denoted as the control (without Biofield Energy Healing Treatment), while the other part was defined as the Biofield Energy Treated sample, which received the Biofield Energy Healing Treatment by renowned Biofield Energy Healer, Alice Branton. Additionally, one group of animals also received Consciousness Energy Healing Treatment per se by the Healer under similar conditions. Vitamin D$_3$ oral formulations were administrated by oral gavage at a dose of 500 µg per kg in groups viz. G1 (untreated Vitamin D$_3$), G2 (Biofield Treated Vitamin D$_3$) and G3 (Biofield Treated animals received untreated Vitamin D$_3$) group. The Biofield Energy Healing Treatment significantly altered the relative oral exposure (AUC$_{0-t}$) of 25-hydroxyvitamin D$_3$ by 54.62% in G2 group compared to the control group, G1. The Biofield Energy Treatment also altered plasma peak concentration (C$_{\text{max}}$) of 25-hydroxyvitamin D$_3$ by 53.8% and 7.6% in G2 and G3 groups, respectively compared to the control group. After oral administration, plasma concentrations of 25-hydroxyvitamin D$_3$ in control group (G1) declined with slow excretion rate and the elimination half-life (T$_{1/2}$) was 100.59 hours. The oral elimination half-life (T$_{1/2}$) of 25-hydroxyvitamin D$_3$ in G2 and G3 were 69.09 hours and 55.76 hours, respectively. The mean residence time (MRT$_{\text{last}}$) and elimination rate constant (K$_{\text{el}}$) remained unaltered in all three groups. The Trivedi Effect®-Energy of Consciousness Healing Treatment could be an innovative strategy which opens new avenues to overcome poorly absorbed pharmaceuticals/nutraceuticals/herbal extracts and can improve the therapeutic performance of orally active molecules.

Keywords: Vitamin D, 25-hydroxyvitamin D$_3$, Ergocalciferol, Cholecalciferol, Biofield Energy Healing Treatment, Pharmacokinetics, Bioavailability, LC-MS/MS

1. Introduction

Vitamin D is a fat soluble essential nutrient and hormone, which plays an important function in order to maintain the healthy immune system and the prevention of diseases [1]. Fortified foods with vitamin D, like dairy products, breakfast cereals, oatmeal, soy milk, and vitamin supplements are the
good sources of vitamin D, while fatty fish such as salmon, mackerel, sardines and tuna, cod liver oil, egg yolks, cheese, etc. are other diet sources. Vitamin D is found naturally in two different forms viz. D$_2$ (ergocalciferol) and D$_3$ (cholecalciferol), while the D$_3$ form is produced by our skin after exposure to the sunlight. Ergocalciferol and cholecalciferol are the biologically inactive pro-hormones [2]. Vitamin D undergoes two biotransformation steps for activation in order to carry out their biological functions. The first transformation occurs in the liver and leads to the formation of 25-hydroxyvitamin D$_3$ [symbol-25(OH) D$_3$; known as calcidiol] via CYP2R1/ CYP27A1 pathway [3]. Thus, the amount of vitamin D$_3$ status obtained from various sources can be best identified in the blood for a relatively long time period [4]. The second biotransformation primarily occurs in the kidney results in the formation of 1 alpha, 25-hydroxyvitamin D$_3$ via CYP27B1 [symbol-1,25(OH)$_2$D$_3$; known as calcitriol] which is the biologically active vitamin D$_3$ [5]. This form is not regarded as a good indicator of vitamin D$_3$ status because before use up it doesn’t last very long in the blood [6]. Vitamin D$_3$ maintains and helps to absorb the calcium and phosphorus level that results in balanced skeletal metabolism and calcium homeostasis [7, 8]. It also plays an important role in maintaining the immunity, cardiovascular, and reproductive systems [9, 10]. Vitamin D and calcium deficiencies are linked and would lead to rickets and osteomalacia. However, its deficiency was also associated with the breast and colorectal cancers, rheumatoid arthritis, multiple sclerosis, Parkinson’s and Alzheimer’s diseases, dementia, and diabetes [11-15]. There are several reports which suggest that most of people Worldwide who are either deficient in (serum concentrations below 20 ng/ml ≈ 50 nmol/L) or have insufficient (20-30 ng/ml ≈ 50 -75 nmol/L) vitamin D$_3$ [16-18], a problem which can be addressed by fortifying foods with vitamin D$_3$ and calcium. The mechanism of transformation of vitamin D and absorption kinetics of active form, vitamin D$_3$ are very complicated to be concluded. Various factors are available that directly affect the vitamin D$_3$ bioavailability such as dietary fiber, genetic factors, and effect of vitamin D$_3$ status [19]. Therefore, it is required to know how vitamin D$_3$ metabolism modifies the active forms of vitamin D$_3$ that circulate in the blood. Therefore, the current study was undertaken to assess the effects of the Biofield Energy Treatment on vitamin D$_3$ bioavailability in rats.

Complementary and Alternate Medicine (CAM) methods have been reported with many clinical beneficial effects in energy healing therapies. Immune system function was significantly improved after Biofield Energy Treatment in case of cervical cancer patients [20], massage therapy [21], etc. Biofield Energy therapy has been discovered thousands year back, which were practiced worldwide such as improved quality of life in case of cancer patient [22], improved functional ability in case of arthritis patient [23], decreased pain and anxiety [24]. National Center for Complementary/Alternative Medicine (NCCAM) has recommended with significant clinical outcome in various clinical pathogenic conditions [25, 26]. Biofield is generated from internal human processes such as blood flow, lymph flow, brain functions, and heart function. This energy can be harnessed and can transmit it into living organism and non-living materials by the process of Biofield Energy Healing. The Trivedi Effect$^\text{a}$-Energy of Consciousness Healing Treatment had been extensively studied in the field of medical science [27, 28], microbiology [29-32], genetics and biotechnology [33, 34], nutraceuticals [35, 36], agricultural science and livestock [37-40], and materials science [41-43].

Recently, it has been reported that The Trivedi Effect$^\text{a}$ has significant capability to alter the physicochemical and thermal properties of various pharmaceuticals, nutraceuticals, and organic compounds through the possible intervention of neutrinos [44-46]. The Trivedi Effect$^\text{a}$-Consciousness Energy Healing Treatment would be a useful approach for the enhancement of the bioavailability of pharmaceuticals and nutraceuticals. Thus, the aim of this study was to evaluate the effect of Biofield Energy Healing Treatment on the plasma pharmacokinetics of 25(OH)D$_3$ in rats after a single oral dose of vitamin D$_3$.

2. Materials and Methods

2.1. Chemicals and Reagents

Vitamin D$_3$, 25-hydroxyvitamin D$_3$ and telmisartan were purchased from Sigma (St. Louis, MO, USA). The reagents used for sample preparation and bioanalysis included acetonitrile (HPLC Grade, Merck), methanol (HPLC Grade, Merck), water (Milli-Q), and formic acid (LC-MS Grade, Fluka). USP grade nitrogen was used as the curtain gas and collision gas for LC-MS/MS were supplied from air compressor (Anesta Iwata, Japan), polypropylene tubes (Tarsons, India), class-A, measuring cylinders and volumetric flasks (Borosiil, Germany) and membrane filters, 0.22 µm and 0.45 µm (Millipore) were used during the study. All other reagents and solvents were of analytical grade available from India.

2.2. Energy of Consciousness Treatment Strategies

The test item, vitamin D$_3$ was divided into two groups, one part was considered as the control sample, while other part of test item was known as the Biofield Energy Treated test sample. The treated test item group was subjected to The Trivedi Effect$^\text{a}$-Energy of Consciousness Healing Treatment by Biofield Energy Healer (also known as The Trivedi Effect$^\text{a}$). The Biofield Energy Treatment was provided by a renowned Biofield Energy Healer, Alice Branton, USA. Moreover, one group of animals also received the Biofield Energy Treatment per se by the same Biofield Energy Healer under similar conditions. This Biofield Treatment was provided for 5 minutes through the Biofield Energy Healer’s unique Energy Transmission process (The Trivedi Effect$^\text{a}$), administered to the test formulation. Similarly, the control formulation was subjected to “sham” heater for 5 minutes, under the same laboratory conditions. The sham heater did
not have any knowledge about the Biofield Energy Treatment. After that, the Biofield Energy Treated and untreated test items were kept in similar sealed conditions and used for the study as per design.

2.3. In Vivo Pharmacokinetics Study

2.3.1. Animals

Male Sprague-Dawley (SD) rats (body weight 230 to 270 grams) were procured from Liveon Biosciences, Bangalore, India. Animals were housed in polycarbonate cage. Temperature and humidity were maintained at 22 ± 3°C and 40-70%, respectively and illumination was controlled to give a sequence of 12 hours light and 12 hours dark cycle. The temperature and humidity were recorded by auto-controlled data logger system. All the animals were provided laboratory rodent diet (Vetcare India Pvt. Ltd, Bengaluru). Reverse osmosis water treated with ultraviolet light was provided ad libitum. The experiments using animals in this investigation were performed in accordance with the guidelines provided by the Committee for the Purpose of Control and Supervision of Experiments on Animals (CPCSEA) as published in The Gazette of India, January 7, 2010 and protocol approved by the Institutional (GVK Bio) Animal Ethics Committee (IAEC approval number: BA-011).

2.3.2. Experimental Design

Rats were divided into three groups (n = 3): group 1 (Gr. 1) – per oral (p.o.) dosing of untreated vitamin D₃, group 2 (Gr. 2) – per oral (p.o.) dosing of Biofield Energy Treated vitamin D₃ and group 3 (Gr. 3) – per oral (p.o.) dosing of untreated vitamin D₃ in Biofield Energy Treated animals. All animals were received per oral dose at 500 g/kg of vitamin D₃ solution formulation. The dose (500 mg/kg) of the test item was chosen based on the preliminary experiments performed in the laboratory and observed the quantifiable concentration of this analyte in rat plasma.

2.4. Formulation Preparation

Solution formulation of the test item was prepared in 14% v/v propylene glycol, 1% v/v Tween 80, 45% v/v PEG, and 40% w/v 2-Hydroxypropyl-β-cyclodextrin in distilled water. All formulations were prepared freshly prior to dosing. The dose volume for per oral route was 5 mL/kg.

2.5. Pharmacokinetic Studies

The solution of test formulations were freshly prepared for per oral dosing. All rats were fasted overnight and the fasting continued up to 4 hours post dosing with free access to drinking water. The oral test formulation was administered at 500 µg/kg dose through oral gavage using an 18G stainless steel intubation cannula. The dosing volume administered was 5 mL/kg. Blood samples (~120 µL) were collected from the jugular vein catheter of three rats from each group at each time point [pre-dose, 0.25, 0.5, 1, 2, 4, 8, 24, 36, 48, 72, and 96 hours (p.o.)]. Samples were collected into labeled micro centrifuge tubes, containing 20% w/v K₃EDTA as an anticoagulant. Plasma samples were separated from the blood by centrifugation at 2500 g for 10 min at 4 ± 2°C and stored below -40°C (Thermo Scientific, USA) deep freezer until bioanalysis.

2.6. LC-MS/MS Analysis

LC-MS/MS analysis of rat plasma samples was performed using API 5500 QTRAP Applied Biosystem/MDS SCIEX (Concord, Ontario, Canada) triple quadruple mass analyzer system with a TurbolonSpray interface connected to a Shimadzu UFLC system (Shimadzu Corp., Japan). The optimum operating parameters were determined by atmospheric pressure chemical ionization (APCI) interface in positive ion mode. A generic mass spectrometry parameters of the analyte were developed and used for the analysis. These parameters were the declustering potential range (80), collision energy range (21), collision cell exit potential range (13), curtain gas (40 arbitrary units), collisionally activated dissociation gas (medium), ionspray voltage (5500 V), source temperature (550°C), and ion source/nebulizer gas 1 (70, arbitrary units each). Interface heaters were kept on for the analyte. The analyte was detected by positive ion spray in the multiple reaction monitoring mode (MRM) mode using predetermined parent/product mass transition ion pairs. The parameters of the selected MRM monitoring transitions for the [M + H]+ precursor ion to selected product ion (m/z) were optimized with 383.20/365.40 (25-hydroxyvitamin D₃), and 515.30/276.20 (telmisartan as an internal standard). The whole system was controlled by Analyst® Classic 1.6.3 software (Applied Biosystem/MDS SCIEX, Concord Canada). Stock solutions of 25-hydroxyvitamin D₃ and telmisartan (internal standard, IS) were prepared in methanol at approximately 2.368 mg/mL and 0.98 mg/mL, respectively and subsequently diluted which were used for the bioanalysis.

The extraction procedure for plasma samples or the spiked into plasma calibration standards were identical. A 50 µL sample of either study sample or spiked calibration standard was added to individual pre-labeled micro-centrifuge tubes. A 50 µL sample of either study sample or spiked calibration standard/quality control samples were added to individual wells of 96 well plate with 500 µL capacity, 200 µL of internal standard (IS) prepared in acetonitrile (ACN) was added to the samples in deep well plate except for blank, where 200 µL of ACN was added and vortexed for 5 minutes. Samples were centrifuged for 10 minutes at a speed of 4000 rpm (3220 g) at 4°C. Following centrifugation, 120 µL of supernatant was transferred into 1000 µL capacity deep well plate and mixed with 120 µL of methanol: water, 50:50 v/v. The plate was kept in the auto-sampler for the LC-MS/MS analysis.

A Shimadzu LC-20AD LC system (Shimadzu Corp., Japan) was connected to a SIL -20 AC HT auto-sampler (Shimadzu Corp., Japan). The supernatant was injected (20 µL) onto a 50 x 4.6 mm (3.5 µm) Waters, X-Bridge, C18 HPLC column (Waters, Massachusetts, Ireland). Analytes were eluted using a gradient elution program with a mobile
phase consists of 0.1% formic acid in water (pump A) with methanol (pump B) at a flow rate of 1.0 mL/min. The column temperature was at 40°C and the sample temperature was at 15°C. The following linear gradient was employed for the separation: 80% A for 0.01 min, 5% A at 2.5 min and hold to 4.5 min, 80% A at 4.9 min, and hold to 6.0 min. The 25-hydroxyvitamin D \_3 and telmisartan elution times were approximately 3.61 and 2.45 min, respectively. Peak integration, regression and calculation of analytes concentration were computed using Analyst Classic (Version 1.6.3) software. The calibration curve was performed by linear curve fit of the peak area ratio (analyte/internal standard) as a function of the concentration in the respective matrix. A weighting of 1/x^2 (where x is the concentration of a given calibration standard level) was found to be optimal. The Lower limit of quantification (LLOQ) in rat plasma was 1.02 ng/mL for 25-hydroxyvitamin D \_3. Analysis of 25-hydroxyvitamin D \_3 in plasma (1.09 to 252.65 ng/mL) showed a repeatability (relative standard deviation-RSD%) of 2.2% to 9.6% and accuracy of 85.70% to 95.65%.

### 2.7. Pharmacokinetic Analysis

The pharmacokinetic parameters of 25-hydroxyvitamin D \_3 were obtained by noncompartmental analysis module in Phoenix WinNonlin® (Version 7.0) (Pharsight, Mountain View, CA). The areas under the concentration time curve (AUC) were obtained by noncompartmental analysis module in Phoenix WinNonlin® (Version 7.0) (Pharsight, Mountain View, CA). The areas under the concentration time curve were computed using Analyst Classic (Version 1.6.3) software. The pharmacokinetic parameters of 25-hydroxyvitamin D \_3 of 5128.77 ng/mL in control group (G1) declined with slow excretion and the elimination half-life (T_{1/2}) was 100.59 hours. The oral elimination half-life (T_{1/2}) was 22.67 ± 14.05 hours. The mean residence time (MAT, mean absorption time; Fr: Relative oral Bioavailability) was 44.14 ± 2.20

The data are expressed as mean values. AUC, area under the plasma concentration–time curve from 0 hours to infinity; CL/F: apparent oral plasma clearance; Vd/F: apparent volume of distribution; C\_max: peak concentration; T\_max: time to reach peak concentration; T\_{1/2}: terminal half-life; K\_el: absorption rate constant; K\_a: absorption rate constant, MRT: mean residence time; MAT: mean absorption time; Fr: Relative oral Bioavailability.

<table>
<thead>
<tr>
<th>Parameter</th>
<th>25-hydroxyvitamin D _3</th>
<th>25-hydroxyvitamin D _3</th>
<th>25-hydroxyvitamin D _3</th>
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</thead>
<tbody>
<tr>
<td>Formulation</td>
<td>Gr. 1-Utreated vitamin D _3</td>
<td>Gr. 2-Biofield Treated vitamin D _3</td>
<td>Gr. 3-Biofield Treated Rats + Untreated vitamin D _3</td>
</tr>
<tr>
<td>C_max (ng/mL)</td>
<td>78.67 ± 21.59</td>
<td>36.34 ± 1.21</td>
<td>72.71 ±17.32</td>
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<tr>
<td>T_max (hours)</td>
<td>22.67 ± 14.05</td>
<td>32.00 ± 6.93</td>
<td>21.33 ± 23.09</td>
</tr>
<tr>
<td>AUC_{0-\infty} (ng/mL*hours)</td>
<td>5128.77 ± 1513.27</td>
<td>2327.56 ± 74.98</td>
<td>4858.98 ± 1330.70</td>
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<td>T_{1/2} (hours)</td>
<td>100.59 ± 57.05</td>
<td>69.09 ± 10.35</td>
<td>55.76 ± 27.74</td>
</tr>
<tr>
<td>MRT_{0-\infty} (hours)</td>
<td>44.36 ± 4.45</td>
<td>45.32 ± 1.47</td>
<td>44.14 ± 2.20</td>
</tr>
<tr>
<td>K_el (hours^-1)</td>
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<td>0.01 ± 0.00</td>
<td>0.01 ± 0.00</td>
</tr>
<tr>
<td>% Change in Fr</td>
<td>54.62</td>
<td>54.62</td>
<td>54.62</td>
</tr>
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The C\_max of 25-hydroxyvitamin D \_3 in control group (G1) was 78.67 ng/mL after 22.67 hours, whereas it was 36.34 ng/mL and 72.71 ng/mL for the 25-hydroxyvitamin D \_3 after 32 hours and 21.33 hours in G2 and G3 group, respectively. The results showed that plasma 25-hydroxyvitamin D \_3 had an oral exposure (AUC\_el) of 5128.77 ng/mL in control (untreated) group. After the Biofield Energy Treatment by a renowned Biofield Energy Healer, Alice Branton, the relative oral exposure (AUC\_el) of 25-hydroxyvitamin D \_3 was altered significantly by 54.62% in G2 group, as compared to the control group. Energy Treatment also altered plasma peak concentration (C\_max) of 25-hydroxyvitamin D \_3 by 53.8% and 7.6% in G2 and G3 groups, respectively as compared to the control group.

After oral administration, plasma concentrations of 25-hydroxyvitamin D \_3 in control group (G1) declined with slow excretion and the elimination half-life (T\_{1/2}) of 25-hydroxyvitamin D \_3 in the Biofield Energy Treatment group (G2) and Biofield Energy Treated rat group (G3) were 69.09 hours and 55.76 hours, respectively. The mean residence time (MRT_{0-\infty}) and elimination rate constant (K\_el) were unaltered in all three groups. These data demonstrates altered plasma exposure and peak plasma levels of 25-hydroxyvitamin D \_3 in rats which might be translated into altered in vivo biological activity of vitamin D \_3. An alteration of oral 25-hydroxyvitamin D \_3 relative bioavailability might be due to the alteration its physicochemical properties and thermal properties by Biofield Energy Treatment.
Figure 1. Mean plasma concentration–time profiles of 25-hydroxyvitamin D$_3$ after per oral (p.o.) administration of vitamin D$_3$ (500 µg/kg) to Sprague Dawley male rats. The data are expressed as mean ± S.D (n = 3).

The results indicated that the Biofield Energy Treated vitamin D$_3$ and animals per se significantly altered the rate and extent of oral absorption of 25-hydroxyvitamin D$_3$. The altered absorption may be due to the alteration of the specific surface area of the vitamin D$_3$ formulation, or the stability of the vitamin D$_3$ formulation in the gastrointestinal tract or due to the altered vitamin D$_3$ metabolism pathways. The significant alteration of relative oral bioavailability of 25-hydroxyvitamin D$_3$ in the Biofield Energy Treated group might be translated into better pharmacological effects in various disease conditions.

It is reported that most people need dietary vitamin D$_3$ to reach the recommended serum level, i.e., greater than 30 ng/mL (≈ 75 nmol/L) [47]. Very few naturally occurring foods contain vitamin D$_3$. Fatty fish flesh and its cod liver oils are the best sources. Small amounts of vitamin D$_3$ are found in beef liver, dairy products, and egg yolk [48, 49]. Vitamin D$_3$ in these foods occurs primarily as vitamin D$_3$ and its metabolite 25(OH)D$_3$. Data suggested that mushrooms have variable amounts of vitamin D$_3$ [50]. In all age groups, e.g., infants, adults, pregnant mothers, elderly are suffering from fat malabsorption, consumption of vitamin D$_3$ supplements or vitamin D$_3$ fortified foods are required to meet the daily need, i.e. approximately 2000 IU/day to maintain serum 25(OH)D$_3$ levels greater than 30 ng/mL [51, 52]. Numerous recent studies suggested that vitamin D$_3$ has the significant roles in bone metabolism and immunity [53, 54]. Vitamin D$_3$ status is inversely associated with the incidence of several diseases, e.g. hyperparathyroidism, cancers, cardio-vascular diseases, diabetes, metabolic disorders, multiple sclerosis, and neurodegenerative diseases [55-58]. Surprisingly, there is very little knowledge on factors that affect absorption and bioavailability of this fat-soluble vitamin. Vitamin D$_3$ deficiency leads to many severe health complications and hence plays an important role in the body. Vitamin D$_3$ helps to improve the calcium absorption in the gut and to maintain normal levels of calcium and phosphates in the blood for bone formation and remodeling [59].

Low plasma exposure of active metabolite(s) of vitamin D$_3$ after a single oral dose of administration in the rat is a key factor that may reduce the efficacy of vitamin D$_3$ [48]. Therefore, this study was carried out to evaluate the pharmacokinetic properties of vitamin D$_3$ using the novel technique known as The Trivedi Effect®-Energy of Consciousness Healing Treatment on vitamin D$_3$. To the authors’ knowledge, this is the first report to demonstrate the effects of Biofield Energy Treatment on vitamin D$_3$ pharmacokinetics in rats after a single dose of oral administration. Pharmacokinetic profiles of 25-hydroxyvitamin D$_3$ in three different groups were compared in male rats following a single oral (gavage) dose of vitamin D$_3$. The study results demonstrated that markedly altered $C_{\text{max}}$ values for the biofield treated groups (G2 and G3) were observed as compared to the control group (G1). The relative oral exposure (AUC$_{0-t}$) of the Biofield Energy Treated group (G2) was significantly altered than the control group (G1). These results clearly demonstrate the significant effect of the Biofield Energy Treatment on the alteration of oral exposure of 25-hydroxyvitamin D$_3$.

4. Conclusions

The Trivedi Effect®-Energy of Consciousness Healing Treatment significantly altered the relative oral exposure (AUC$_{0-t}$) of 25-hydroxyvitamin D$_3$ by 54.62% in G2 group compared to the control group, G1. The Biofield Energy Treatment also altered plasma peak concentration (C$_{\text{max}}$) of 25-hydroxyvitamin D$_3$ by 53.8% and 7.6% in G2 and G3 groups, respectively as compared to the control group. After oral administration, plasma concentrations of 25-hydroxyvitamin D$_3$ in the control group (G1) declined with slow excretion and the elimination half-life (T$_{1/2}$) was 100.59 hours. The oral elimination half-life (T$_{1/2}$) of 25-
hydroxyvitamin D$_3$ in the Biofield Energy Treatment group (G2) and Biofield Energy Treated rat group (G3) were 69.09 hours and 55.76 hours, respectively. The mean residence time (MRT$_{\text{res}}$) and elimination rate constant (K$_{\text{e}}$) were unaltered in all three groups. These data demonstrates altered plasma exposure and peak plasma levels of 25-hydroxyvitamin D$_3$ in rats which might be translated into altered in vivo biological activity of vitamin D$_3$. An alteration of oral exposure of 25-hydroxyvitamin D$_3$ relative might be due to the alteration its physicochemical properties and thermal properties by Biofield Energy Treatment. Hence, The Trivedi Effect$^5$-Energy of Consciousness Healing Treatment is considered as an innovative strategy which opens new opportunities in order to improve the poorly absorbed drug/nutraceuticals/herbal extracts that can improve the activity of vitamin D$_3$. As a result, this treatment might be beneficial to the different groups of cardiac and kidney transplant patients, hip fracture patients, osteoporotic patients, hyperparathyroidism and cancer patient, neurodegenerative and ischemic heart patients.

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