

# Journal of Analytical, Bioanalytical and Separation Techniques

ISSN: 2476-1869

Research Article



OPEN ACCESS

DOI: 10.15436/2476-1869.19.3179

## Evaluation of the Structural Properties and Isotopic Abundance Ratios of Consciousness Energy Healing Treated Vitamin D<sub>3</sub> Using LC-MS, GC-MS, and NMR Spectroscopy

Mahendra Kumar Trivedi<sup>1</sup>, Snehasis Jana<sup>2\*</sup>

<sup>1</sup>Trivedi Global, Inc., Henderson, Nevada, USA

<sup>2</sup>Trivedi Science Research Laboratory Pvt. Ltd., Thane-West, Maharashtra, India

\*Corresponding author: Dr. Snehasis Jana, Trivedi Science Research Laboratory Pvt. Ltd., Thane-West, Maharashtra, India; E-mail: publication@trivedisrl.com

### Abstract

Vitamin D<sub>3</sub> (cholecalciferol) has multiple effects which regulate the functions of muscles, brain, lungs, liver, kidneys, heart, immune system, pancreas, large and small intestines. The aim of the study was to investigate the impact of The Trivedi Effect<sup>®</sup>-Consciousness Energy Healing Treatment on the isotopic abundance ratios ( $P_{M+1}/P_M$  and  $P_{M+2}/P_M$ ) and structural properties of vitamin D<sub>3</sub> using LC-MS, GC-MS, and NMR spectroscopy. Vitamin D<sub>3</sub> was divided into two parts, one part of the sample was termed as untreated (control) sample, while the other part of the sample received The Trivedi Effect<sup>®</sup>-Biofield Energy Healing Treatment remotely by a renowned Biofield Energy Healer, Mr. Mahendra Kumar Trivedi and termed as Biofield Energy Treated sample. The LC-ESI-MS analysis of both the samples showed the mass of protonated ion at  $m/z$  385.3 (calcd for  $C_{27}H_{45}O^+$ , 385.35) at the retention time ( $R_t$ ) 20.7 minutes. The liquid chromatographic peak area of the treated sample was significantly improved by 16.00% compared to the control sample. Similarly, the isotopic abundance ratio of  $P_{M+1}/P_M$  ( $^2H/^1H$  or  $^{13}C/^12C$  or  $^{17}O/^16O$ ) was very significantly increased by 470.88% in the treated sample compared to the control sample. But, the isotopic abundance ratio of  $P_{M+2}/P_M$  ( $^{18}O/^16O$ ) in the treated sample also significantly decreased by 41.81% compared to the control sample. Therefore, the  $^2H$ ,  $^{13}C$ , and  $^{17}O$  contributions from  $C_{27}H_{45}O^+$  to  $m/z$  386 were significantly increased, whereas  $^{18}O$  contribution from  $C_{21}H_{21}O_6^+$  to  $m/z$  387 was decreased in the treated sample compared with the control sample. The GC-MS analysis showed that the parent molecular mass peak intensities ( $m/z$  384.4) in the treated sample at  $R_t$  23.04 and 23.65 minutes were significantly increased by 6.11% and 9.62%, respectively compared with the control sample. The proton and carbon signals for  $CH_3$ ,  $CH_2$ ,  $CH$ ,  $C-OH$ , and  $=C=$  groups in the  $^1H$  and  $^{13}C$  NMR spectra of the control and treated samples were similar. The altered isotopic abundance ratios and mass peak intensities might be due to energy produced by The Trivedi Effect<sup>®</sup>-Consciousness Energy Healing Treatment *via* the possible mediation of neutrinos, which further lead to the change in the kinetic isotope effects and physicochemical properties of the treated sample. Thus, The Trivedi Effect<sup>®</sup> Treated vitamin D<sub>3</sub> would be advantageous for designing better nutraceutical and pharmaceutical formulations which might provide better therapeutic response against vitamin D deficiency, rickets, osteoporosis, arthritis, multiple sclerosis, diabetes, cancer, cardiovascular diseases, mental disorders, infections, stress, aging, glucose intolerance, multiple sclerosis, Parkinson's and Alzheimer's diseases, dementia, *etc.*

**Keywords:** Vitamin D<sub>3</sub>; The Trivedi Effect<sup>®</sup>; Energy of Consciousness Healing Treatment; LC-MS; Isotopic abundance; GC-MS; Kinetic isotope effects

Received date: January 19, 2021

Accepted date: February 19, 2021

Published date: February 25, 2021

**Citation:** Trivedi, MK., et al. Evaluation of the Structural Properties and Isotopic Abundance Ratios of Consciousness Energy Healing Treated Vitamin D<sub>3</sub> Using LC-MS, GC-MS, and NMR Spectroscopy. (2021) J Anal Bioanal Separat Techq 5(1): 1-8.

**Copyright:** © 2021 Trivedi MK. This is an Open access article distributed under the terms of Creative Commons Attribution 4.0 International License.

## Introduction

Vitamin D<sub>3</sub> is a fat soluble vitamin, scientifically also known as cholecalciferol, found in foods like cod liver oil, mushrooms (if exposed to ultraviolet light), mackerel, halibut, canned sock-eye salmon and also in the dietary supplements<sup>[1-3]</sup>. Vitamin D has multiple roles in the human body, which regulate the functions of muscles, brain, lungs, liver, kidneys, heart, immune system, pancreas, large and small intestines. Vitamin D receptors are ubiquitously present in most of the body organs, *i.e.*, brain, heart, lungs, kidney, liver, pancreas, large and small intestines, muscles, reproductive, nervous system, *etc.* Vitamin D receptor response elements with hundreds of genes directly or indirectly influence cell-to-cell communication, normal cell growth, cell differentiation, cell cycling and proliferation, hormonal balance, maintenance of calcium and phosphorus balance, neurotransmission, skin health, immune and cardiovascular function<sup>[1-3]</sup>. Insufficient dietary intake, intestinal malabsorption or chronic liver disease, familial hypophosphatemia, hypocalcaemia that is associated with hypoparathyroidism, and those who fail to produce enough vitamin D<sub>3</sub> in their skin from its precursor, 7-dehydrocholesterol, in response to exposure to ultraviolet light lead to vitamin D deficiency. Vitamin D deficiency responsible for several diseases, e.g., rickets, osteoporosis, arthritis, multiple sclerosis, cancer, diabetes mellitus, mental disorders, cardiovascular diseases, inflammations, infections, stress, aging, cognitive impairment in older adults, glucose intolerance, multiple sclerosis, Parkinson's and Alzheimer's diseases, dementia, *etc.*<sup>[1-3,4-6]</sup>. The National Academies Press (USA) report suggested that 15 µg/d (600 IU per day) of vitamin D is required for all individuals (males, female, pregnant/lactating women) between the ages of 1 and 70 years old<sup>[7]</sup>. The toxicity like hypercalcemia, polyuria, polydipsia, weakness, mental retardation, and insomnia may be likely to cause due to the higher dose of vitamin D supplementation<sup>[8]</sup>. Vitamin D<sub>3</sub> is highly heat and light-sensitive compound<sup>[9,10]</sup>. Therefore, the stability of vitamin D<sub>3</sub> is more concerned. The mechanism of transformation of vitamin D and its absorption kinetics of active form (vitamin D<sub>3</sub>) is very complicated. The bioavailability absorption and bioavailability of vitamin D<sub>3</sub> directly affected by various factors such as dietary fiber, genetic factors, and status of vitamin D<sub>3</sub><sup>[11]</sup>.

Some of the recent studies revealed that The Trivedi Effect®-Energy of Consciousness Healing Treatment significantly improved the bioavailability of various pharmaceutical/nutraceutical compounds, *i.e.*, resveratrol, berberine, and 25-hydroxyvitamin D<sub>3</sub> in Male Sprague-Dawley rats<sup>[12-14]</sup>. Biofield is the electromagnetic field exist around the human body<sup>[15,16]</sup>. The Trivedi Effect® is a natural and only scientifically proven phenomenon in which a person can harness this inherently intelligent energy (Universal Energy Field) and transmit it anywhere on the planet. Several Biofield based Energy Healing Therapies used against various human disease conditions<sup>[17,18]</sup>. Biofield Energy Healing therapy has been recognized worldwide as a Complementary and Alternative Medicine (CAM) health care approach by National Center of Complementary and Integrative Health (NCCIH) with other therapies, medicines and practices such as Ayurvedic medicine, traditional Chinese herbs and medicines, aromatherapy, yoga, Tai Chi, Qi Gong, chiropractic/osteopathic manipulation, meditation, homeopathy, acupuncture,

acupressure, healing touch, hypnotherapy, movement therapy, naturopathy, cranial sacral therapy, Reiki, *etc.*<sup>[19]</sup>. The Trivedi Effect®-Energy of Consciousness Healing Treatment has astonishing ability to transform the characteristic properties of organic compounds<sup>[20,21]</sup>, metals and ceramic<sup>[22,23]</sup>, nutraceuticals<sup>[24,25]</sup>, pharmaceuticals<sup>[26,27]</sup>, productivity of crops<sup>[28,29]</sup>, genetic materials<sup>[30,31]</sup> and alteration of the isotopic abundance ratio in the organic compounds<sup>[32,33]</sup> may be through the possible mediation of neutrinos<sup>[15]</sup>. Several scientific organization is doing extensive research on the natural stable isotopes to understand the isotope effects resulting from the alterations of the isotopic composition, which have lots of implementation in different fields of sciences<sup>[34-36]</sup>. The conventional Gas chromatography – mass spectrometry (GC-MS) and liquid chromatography – mass spectrometry (LC-MS) techniques are widely used for the study of isotope ratio analysis with sufficient precision<sup>[35]</sup>. Considering the various importance of The Trivedi Effect®-Consciousness Energy Healing Treatment and its outstanding impact on various object(s), the isotopic abundance ratio analysis of P<sub>M+1</sub>/P<sub>M</sub> (<sup>2</sup>H/<sup>1</sup>H or <sup>13</sup>C/<sup>12</sup>C or <sup>17</sup>O/<sup>16</sup>O) and P<sub>M+2</sub>/P<sub>M</sub> (<sup>18</sup>O/<sup>16</sup>O) in vitamin D<sub>3</sub> was performed to evaluate the influence of The Trivedi Effect® on the isotopic abundance ratio and structural properties using the sophisticated LC-MS, GC-MS, and NMR (Nuclear Magnetic Resonance) analytical techniques.

## Materials and Methods

### Chemicals and Reagents

Vitamin D<sub>3</sub> (>98%) was purchased from Sigma-Aldrich, India. All other chemicals used during the experiments were of analytical grade available in India.

### Consciousness Energy Healing Treatment Strategies

Vitamin D<sub>3</sub> sample was divided into two parts. One part of vitamin D<sub>3</sub> was considered as control/untreated which was not subjected to the Biofield Energy Treatment. Whereas, the second part of vitamin D<sub>3</sub> sample was treated with The Trivedi Effect®-Energy of Consciousness Healing Treatment remotely under standard laboratory conditions for 3 minutes by a renowned Biofield Energy Healer, Mr. Mahendra Kumar Trivedi (USA) and termed as Biofield Energy Treated sample. This Biofield Energy Treatment was provided through the healer's unique energy transmission process to the test item. Further, the control sample was treated with "sham" healer for the purpose of better comparison. The sham healer did not have any knowledge about the Biofield Energy Treatment. After all, the Biofield Energy Treated and untreated samples of vitamin D<sub>3</sub> were kept in sealed conditions and characterized using LC-MS, GC-MS, and NMR techniques.

### Characterization

**Liquid chromatography-mass spectrometry (LC-MS) analysis and Calculation of Isotopic Abundance Ratio:** The LC-MS analysis of the control and Biofield Energy Treated vitamin D<sub>3</sub> was carried out with the help of LC-Dionex Ultimate 3000, MS-TSQ Endura, USA equipped with a photo-diode array (PDA) detector connected with a triple-stage quadrupole mass spectrometer (Thermo Scientific TSQ Endura, USA) with a Thermo Scientific Ion Max NG source and Atmospheric Pressure

chemical ionization (APCI). The analysis was performed on a reversed phase Zorbax SB-C18 100 × 4.6 mm, 3.5 μm in gradient mode in the liquid chromatograph. The mobile phase was 2mM ammonium formate and 0.5% formic acid in water and acetonitrile at a constant flow rate of 0.6 mL/min. The column temperature was kept constant at 40°C. The injection volume was 10 μL and the total run time was 30 min. Chromatographic separation was achieved using gradient condition as follow: 0 min-50%B, 5 min-90%B, 10 min-100%B, 20 min-100%B, 25 min-50%B, and 30 min-50%B. Peaks were monitored using the PDA detector. Mass spectrometric analysis was performed under +ve ESI mode. The total ion chromatogram, peak area% and mass spectrum of the individual peak which was appeared in LC along with the full scan were recorded. The mass peak intensities of the mass spectrum of the individual peak were recorded.

The natural abundance of C, O, and H isotope can be predicted from the comparison of the relative abundance of the isotope peak with respect to the base peak. The values of the natural isotopic abundance of the common elements are obtained from the literature<sup>[37-40]</sup>. The isotopic abundance ratios ( $P_{M+1}/P_M$  and  $P_{M+2}/P_M$ ) for the control and Biofield Energy Treated vitamin D<sub>3</sub> were calculated.

Percentage (%) change in isotopic abundance ratio of vitamin D<sub>3</sub>  
 $= [(IAR_{Treated} - IAR_{Control}) / IAR_{Control}] \times 100]$

Where,  $IAR_{Treated}$ : isotopic abundance ratio in the treated vitamin D<sub>3</sub> and  $IAR_{Control}$ : isotopic abundance ratio in the control vitamin D<sub>3</sub>.

#### Gas chromatography-mass spectrometry (GC-MS) analysis:

Agilent 7890B Gas chromatograph equipped with a silica capillary column HP-5 MS(30 m x 0.25 mm x 0.25 μm) and coupled to a quadrupole detector with pre-filter (5977B, USA) was operated with electron impact (EI) ionization in positive ion mode at 70 eV. Oven temperature was programmed from 50°C (1 min hold) to 150°C@20°C/min to 200°C (6 min hold)@25°C/min to 280°C@20°C/min(12 min hold). Temperatures of the injector, detector (FID), auxiliary, ion source, and quadrupole detector were 230, 250, 280, 230, and 150°C. Vitamin D<sub>3</sub> was dissolved in methanol, and 5.0 μL was splitlessly injected with helium as a carrier gas with a flow rate of 2.0 mL/min. Mass spectra were scanned from  $m/z$  40 to 1050 at a stability of ± 0.1  $m/z$  mass accuracy over 48 hours and mass peak intensities of the mass spectrum of the individual peak were recorded.

Percent change in peak intensity (I) was calculated using following equations:

Percent change in peak intensity (I) =  $[(I_{Treated} - I_{Control}) / I_{Control}] \times 100$

Where,  $I_{Control}$  and  $I_{Treated}$  are the peak intensity of the control and Biofield Energy Treated samples, respectively.

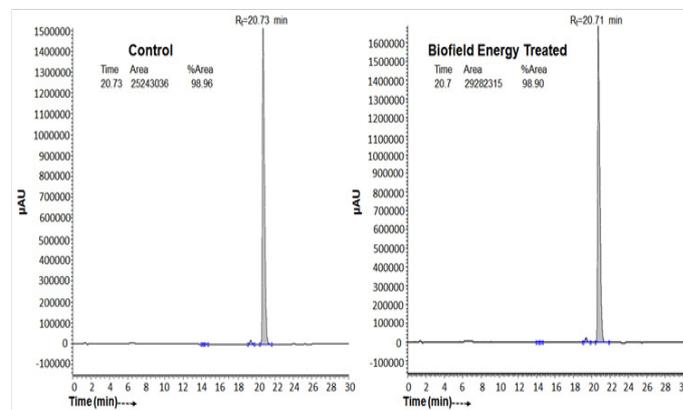
**Nuclear Magnetic Resonance (NMR) Analysis:** <sup>1</sup>H NMR spectra of vitamin D<sub>3</sub> were recorded at 400 MHz on Agilent-MRDD2 FT-NMR. Approximately 3 mg of the sample was dissolved in DMSO-d<sub>6</sub>. Chemical shifts (d) were in parts per million (ppm) relative to the solvent's residual proton chemical shift {(CD<sub>3</sub>)-

<sub>2</sub>SO, δ = 2.5}. <sup>1</sup>H NMR multiplicities were designated as singlet (s), doublet (d), doublet of doublet (dd), triplet (t), quartet (q), multiplet (m), broad (br), apparent (app). Similarly, <sup>13</sup>C NMR spectra of vitamin D<sub>3</sub> were measured at 100 MHz on Agilent-MRDD2 FT-NMR spectrometer at room temperature. Approximately 25 mg of the sample was dissolved in DMSO-d<sub>6</sub>. Chemical shifts (d) were in parts per million (ppm) relative to the solvent's residual carbon chemical shift {(CD<sub>3</sub>)<sub>2</sub>SO, δ = 39.52}.

## Results and Discussion

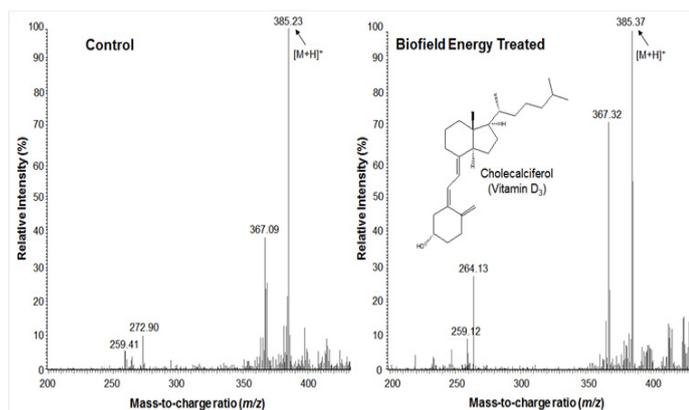
### Liquid Chromatography-Mass Spectrometry (LC-MS) Analysis

The control and Biofield Energy Treated vitamin D<sub>3</sub> showed a clear and sharp chromatographic peak at retention times (Rt) 20.73 and 20.71 minutes, respectively (Figure 1). The peak area of the control and Biofield Energy Treated samples at R<sub>t</sub> 20.7 minutes was 25243036 and 29282315 in control and Biofield Energy Treated sample, respectively. The peak area of the Biofield Energy Treated sample was significantly improved by 16.00% compared to the control sample. It indicated that the polarity of both the samples remained similar, but the solubility profile of the Biofield Energy Treated sample was significantly improved compared to the control sample. Solubility also plays a major role in the dosage forms of parenteral formulations and one of the important parameters to achieve the improved concentration of drug in systemic circulation for achieving elevated pharmacological response<sup>[41]</sup>.



**Figure 1:** Total ion chromatograms (TIC) of the control and Biofield Energy Treated vitamin D<sub>3</sub>.

The ESI-MS mass spectra of the control and Biofield Energy Treated samples at R<sub>t</sub> of 20.7 minutes exhibited the molecular ion peak (Figure 2) of vitamin D<sub>3</sub>(C<sub>27</sub>H<sub>45</sub>O<sup>+</sup>) adduct with hydrogen ion at  $m/z$  385.23 and 385.37, respectively (calcd for C<sub>27</sub>H<sub>45</sub>O<sup>+</sup>, 385.35). The major fragmented mass peak [M-OH]<sup>+</sup> at  $m/z$  367.09 and 367.32 (calcd for C<sub>27</sub>H<sub>43</sub><sup>+</sup>, 367.3) in control and Biofield Energy Treated vitamin D<sub>3</sub>, respectively (Figure 2).



**Figure 2:** The ESI-MS spectra of the control and Biofield Energy Treated vitamin D<sub>3</sub> at R<sub>t</sub> 20.7 minutes in the chromatograms.

The mass fragmentation pattern of the control and Biofield Energy Treated vitamin D<sub>3</sub> in the spectra were the similar type (Figure 2). The parent mass peak at *m/z* 385.3 was the base peak which exhibited 100% relative peak intensity in both the spectra (Figure 2). But, the relative peak intensities of the other ion peaks in the treated vitamin D<sub>3</sub> were significantly altered compared to the control sample.

**Isotopic Abundance Ratio Analysis:** The control and treated samples of vitamin D<sub>3</sub> showed the mass of a protonated molecular ion at *m/z* 385.3 (calcd for C<sub>27</sub>H<sub>45</sub>O<sup>+</sup>, 385.35) with 100% relative abundance in the mass spectra. The theoretical calculation of isotopic peak P<sub>M+1</sub> for the protonated vitamin D<sub>3</sub> presented below:

$$P(^{13}\text{C}) = [(27 \times 1.1\%) \times 100\% \text{ (the actual size of the } M^+ \text{ peak)}] / 100\% = 29.7\%$$

$$P(^2\text{H}) = [(45 \times 0.015\%) \times 100\%] / 100\% = 0.675\%$$

$$P(^{17}\text{O}) = [(1 \times 0.04\%) \times 100\%] / 100\% = 0.04\%$$

$$P_{M+1} \text{ i.e. } ^{13}\text{C}, ^2\text{H}, \text{ and } ^{17}\text{O} \text{ contributions from } C_{27}H_{45}O^+ \text{ to } m/z \text{ 386} = 30.42\%$$

Similarly, the theoretical calculation of isotopic peak P<sub>M+2</sub> for the protonated vitamin D<sub>3</sub> presented below:

$$P(^{18}\text{O}) = [(1 \times 0.20\%) \times 100\%] / 100\% = 0.2\%$$

$$P_{M+2} \text{ of } ^{18}\text{O} \text{ contribution from } C_{27}H_{45}O^+ \text{ to } m/z \text{ 387} = 0.2\%$$

The calculated isotopic abundance of P<sub>M+1</sub> value 30.42% was higher to the observed value (9.72%). But, the calculated P<sub>M+2</sub> value 0.2% was lower to the observed value (12.03%) (Table 1). The probability of A + 1 and A + 2 elements having an isotope with one and two mass unit heavier, respectively than the most abundant isotope (i.e., <sup>13</sup>C, <sup>2</sup>H, <sup>17</sup>O, and <sup>18</sup>O) contributions to the mass of the isotopic molecular ion [M+1]<sup>+</sup> and [M+2]<sup>+</sup>. <sup>2</sup>H did not contribute much any isotopic *m/z* ratios because of its less natural abundance compared to the abundances of C and O isotopes in nature<sup>[38,39]</sup>. But, the contributions of <sup>13</sup>C, <sup>17</sup>O, and <sup>18</sup>O was major from vitamin D<sub>3</sub> to the isotopic mass peak at *m/z*

386 and 387 confirmed from the calculations. Therefore, P<sub>M</sub>, P<sub>M+1</sub>, and P<sub>M+2</sub> of the vitamin D<sub>3</sub> at *m/z* 385, 386, and 387 of the control and Biofield Energy Treated samples were obtained from the experimental relative abundance of M<sup>+</sup>, (M+1)<sup>+</sup>, and (M+2)<sup>+</sup> peaks, respectively in the mass spectra (Table 1).

**Table 1:** LC-ESI-MS isotopic abundance ratio analysis of control and Biofield Energy Treated vitamin D<sub>3</sub>

Parameter	Control sample	Biofield Energy Treated sample
P <sub>M</sub> at <i>m/z</i> 385 (%)	100	100
P <sub>M+1</sub> at <i>m/z</i> 386 (%)	9.72	55.49
P <sub>M+1</sub> /P <sub>M</sub>	0.0972	0.5549
% Change of isotopic abundance ratio (P <sub>M+1</sub> /P <sub>M</sub> ) with respect to the control vitamin D <sub>3</sub>		470.88
P <sub>M+2</sub> at <i>m/z</i> 387 (%)	12.03	7.00
P <sub>M+2</sub> /P <sub>M</sub>	0.1203	0.070
% Change of isotopic abundance ratio (P <sub>M+2</sub> /P <sub>M</sub> ) with respect to the control vitamin D <sub>3</sub>		-41.81

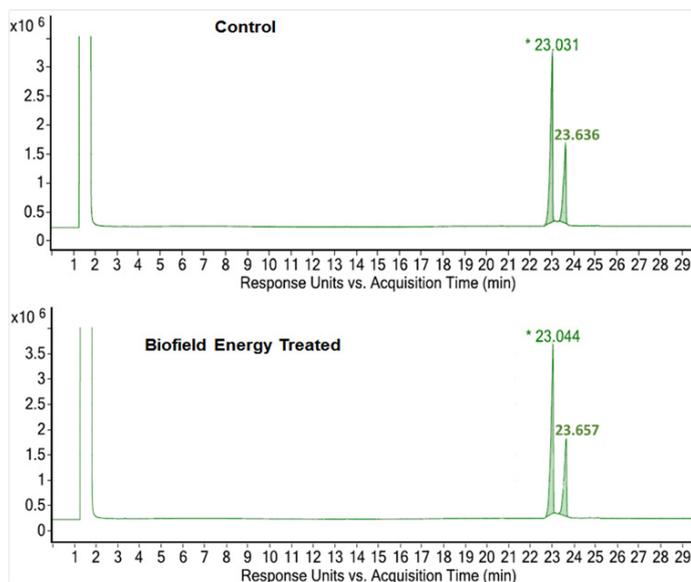
P<sub>M</sub> = the relative peak intensity of the parent molecular ion M<sup>+</sup>; P<sub>M+1</sub> = the relative peak intensity of the isotopic molecular ion [M+1]<sup>+</sup>; P<sub>M+2</sub> = the relative peak intensity of the isotopic molecular ion [M+2]<sup>+</sup>, and M = mass of the parent vitamin D<sub>3</sub> molecule.

The isotopic abundance ratio of P<sub>M+1</sub>/P<sub>M</sub> (<sup>2</sup>H/<sup>1</sup>H or <sup>13</sup>C/<sup>12</sup>C or <sup>17</sup>O/<sup>16</sup>O) in treated vitamin D<sub>3</sub> was very significantly increased by 470.88% compared to the control sample (Table 1). This indicated that the <sup>13</sup>C, <sup>2</sup>H, and <sup>17</sup>O contributions from C<sub>27</sub>H<sub>45</sub>O<sup>+</sup> to the isotopic *m/z* 386 in the Biofield Energy Treated vitamin D<sub>3</sub> sample was very significantly increased compared to the control sample. But, the isotopic abundance ratio of P<sub>M+2</sub>/P<sub>M</sub> (<sup>18</sup>O/<sup>16</sup>O) in the Biofield Energy Treated vitamin D<sub>3</sub> was significantly decreased by 41.81% compared to the control sample (Table 1). Therefore, the <sup>18</sup>O contribution from C<sub>27</sub>H<sub>45</sub>O<sup>+</sup> to the isotopic *m/z* 387 in the Biofield Energy Treated vitamin D<sub>3</sub> was significantly decreased compared with the control sample. Table 1

The recent discovery of neutrino oscillations seems to give credence to the postulates of Mr. Mahendra Kumar Trivedi on The Trivedi Effect<sup>®</sup><sup>[17]</sup>. Neutrinos are one of the most abundant particles in the Universe; however, are very difficult to observe. Neutrino oscillations imply that neutrinos have small but non-zero masses, which have profound implications to our understanding of elementary particle physics and the Universe. Neutrons and alteration in its number in a molecule lead to the increased or decreased isotopic abundance of the compounds. Changes in atomic/molecular weights are postulated to the changes in atomic mass and charge through the possible mediation of neutrinos<sup>[15,42-44]</sup>. Neutrinos only interact *via* the weak force, which is indeed very weak and the consequence is significant. If a neutrino is produced, it travels straightly in any matter as if it is traveling in the vacuum and seldom interacts with matter, i.e., a neutrino produced in the atmosphere of Earth can easily travel through the whole Earth<sup>[42]</sup>. Thus, it can be assumed that The Trivedi Effect<sup>®</sup>-Consciousness Energy Healing Treatment might provide the necessary energy for the neutrino

oscillations leads to the alteration of the fundamental physico-chemical properties of a compound by the interactions to the subject(s)<sup>[45,46]</sup>. The change in the isotopic abundance ratio of the atoms/molecules leads to the change in the kinetic isotope effects, which is very useful to study the reaction mechanism, understand the enzymatic transition state, and enzyme mechanism that is supportive for designing effective and specific inhibitors, *etc*<sup>[36]</sup>. Therefore, The Biofield Energy Treated vitamin D<sub>3</sub> with altered isotopic abundance ratio ( $P_{M+1}/P_M$  and  $P_{M+2}/P_M$ ) would be advantageous for the designing of better nutraceutical and pharmaceutical formulations.

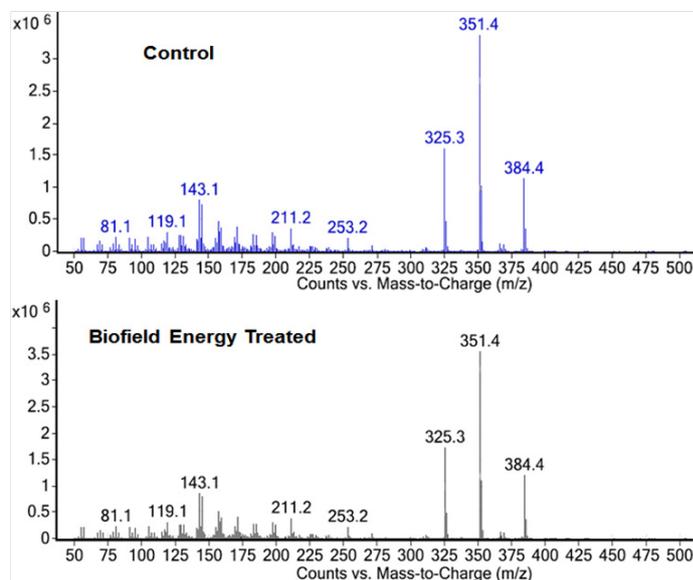
**Gas Chromatography-Mass Spectrometry (GC-MS) Analysis:** The GC chromatograms of vitamin D<sub>3</sub> showed two clear independent peaks both in control, and Biofield Energy Treated samples (Figure 3). The  $R_t$  of the control sample was found at 23.03 and 23.64 minutes, whereas Biofield Energy Treated sample at 23.04 and 23.66 minutes. The results indicated that the polarity of the Biofield Energy Treated sample remained close compared to the control sample. May be due to the *cis* and *trans* isomers of vitamin D<sub>3</sub> both the control and Biofield Energy Treated sample showed two peaks the chromatograms<sup>[47,48]</sup>.



**Figure 3:** GC chromatograms of the control and Biofield Energy Treated vitamin D<sub>3</sub>.

The mass spectra of the control and Biofield Energy Treated vitamin D<sub>3</sub> at  $R_t$  of 23.04 minutes exhibited the presence of the molecular ion ( $C_{27}H_{44}O^+$ ) (Figure 4) at  $m/z$  384.4 (calcd for  $C_{27}H_{44}O^+$ , 384.34). The other mass fragmentation peak at lower  $m/z$  351.4, and 325.3 for  $C_{26}H_{39}^{++}$ , and  $C_{24}H_{37}^+$ , respectively in both the control and Biofield Energy Treated vitamin D<sub>3</sub> (Figure 4). The mass fragmentation pattern of both the samples was similar. But the mass peak intensities of the Biofield Energy Treated vitamin D<sub>3</sub> were significantly altered compared to the control sample. The mass peak intensity of the control and Biofield Energy Treated vitamin D<sub>3</sub> were 1127387.63 and 1196286.75, respectively at  $R_t$  of 23.04 minutes. Similarly, the mass peak intensities of the control and Biofield Energy Treated vitamin D<sub>3</sub> were 545957.06 and 598460.19, respectively at  $R_t$  of 23.65 minutes. The mass peak intensities at  $R_t$  23.04 and 23.65 minutes were significantly increased in the Biofield Energy Treated

sample by 6.11% and 9.62%, respectively (Table 2) compared to the control sample. The mass peak intensities were significantly increased which might be due to the impact of Consciousness Energy Healing Treatment (The Trivedi Effect<sup>®</sup>)<sup>[15,45,46]</sup>. Fig 3, 4 table 2



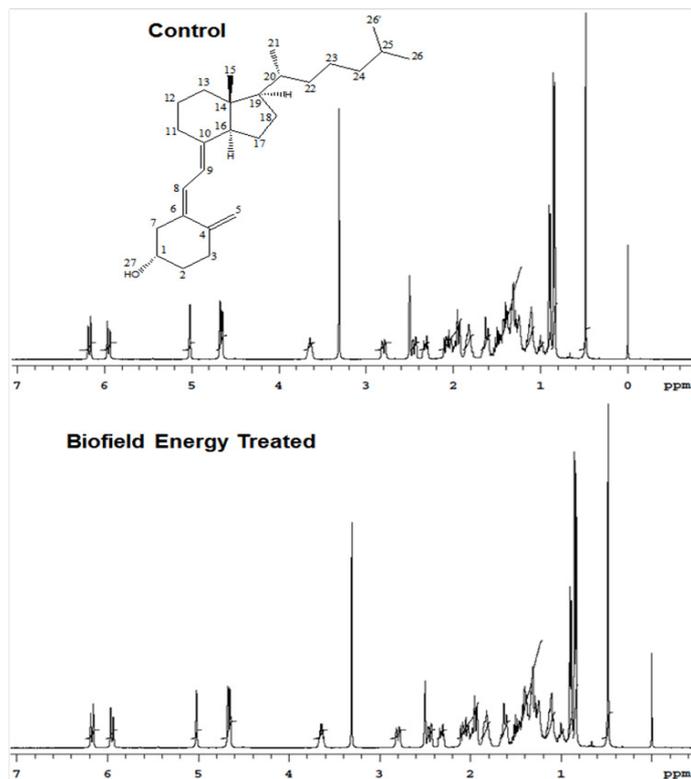
**Figure 4:** GC-MS spectra of the control and Biofield Energy Treated vitamin D<sub>3</sub> at  $R_t$  23.04 minutes.

**Table 2:** GC-MS chromatographic and mass spectra analysis at  $R_t$  23.04 and 23.67 minutes of the control and Biofield Energy Treated vitamin D<sub>3</sub>.

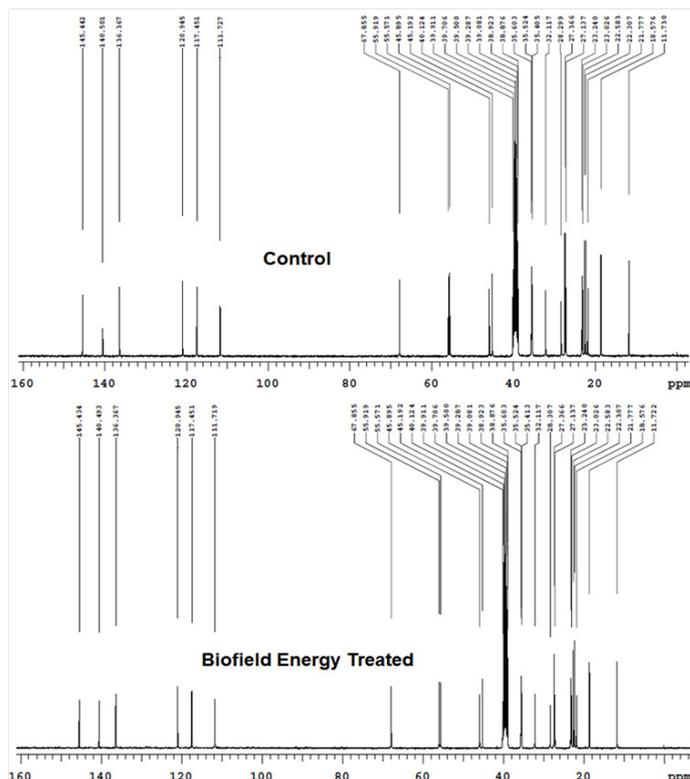
Parameters	Control sample	Biofield Energy Treated sample	% Change
Masspeak ( $m/z=384$ ) intensity at $R_t$ 23.04 minutes	1127387.63	1196286.75	6.11
Mass peak ( $m/z=384$ ) intensity at $R_t$ 23.65 minutes	545957.06	598460.19	9.62

#### Nuclear Magnetic Resonance (NMR) Spectroscopy Analysis:

The <sup>1</sup>H and <sup>13</sup>C NMR spectra shown in Figures 5 and 6, respectively for the control and Biofield Energy Treated vitamin D<sub>3</sub>. The analyzed <sup>1</sup>H and <sup>13</sup>C NMR spectral information are presented in Table 3. The signals for the protons coupling of CH<sub>3</sub>, CH<sub>2</sub>, CH, and OH protons in both the <sup>1</sup>H NMR spectra of vitamin D<sub>3</sub> were in the range of  $\delta$ 0.48 to 6.17 ppm (Figure 5 and Table 3), which were very close to each other. Similarly, the carbon signals for CH<sub>3</sub>, CH<sub>2</sub>, CH, =C=, and C-OH groups in both the <sup>13</sup>C NMR spectra were in the range of 11.72-145.44 (Figure 6 and Table 3). The experimental results were closely matched to the published literature<sup>[48]</sup>. The <sup>1</sup>H and <sup>13</sup>C NMR spectral data indicated that there was no structural modification of The Trivedi Effect<sup>®</sup>-Biofield Energy Treated vitamin D<sub>3</sub> compared to the control sample. Table 3, fig 5, fig 6



**Figure 5:** The <sup>1</sup>H NMR spectra of the control and Biofield Energy Treated vitamin D<sub>3</sub>.



**Figure 6:** The <sup>13</sup>C NMR spectra of the control and Biofield Energy Treated vitamin D<sub>3</sub>.

**Table 3:** <sup>1</sup>H and <sup>13</sup>C NMR spectroscopic data of both the control and Biofield Energy Treated vitamin D<sub>3</sub>

<sup>1</sup> H & <sup>13</sup> C S. No	<sup>1</sup> H NMR δ (ppm) & Multiplicity		<sup>13</sup> C NMR δ (ppm)	
	Untreated	Biofield Energy Treated	Untreated	Biofield Energy Treated
1	3.65 (m, <i>J</i> = 20 Hz, H)	3.65 (m, <i>J</i> = 20 Hz, H)	67.86	67.86
2	1.63 (m, <i>J</i> = 24 Hz, 2H)	1.63 (m, <i>J</i> = 24 Hz, 2H)	35.41	35.41
3	1.82 (m, <i>J</i> = 28 Hz, 2H)	1.82 (m, <i>J</i> = 28 Hz, 2H)	32.12	32.12
4	--	--	136.37	136.37
5	4.67 (d, <i>J</i> = 12 Hz, 2H)	4.67 (d, <i>J</i> = 12 Hz, 2H)	111.73	111.72
6	--	--	145.44	145.43
7	1.93-2.11 (m, 2H)	1.93-2.11 (m, 2H)	45.90	45.89
8	6.17 (d, <i>J</i> = 12 Hz, H)	6.17 (d, <i>J</i> = 12 Hz, H)	120.95	120.95
9	5.95 (d, <i>J</i> = 12 Hz, H)	5.95 (d, <i>J</i> = 12 Hz, H)	117.45	117.45
10	--	--	140.51	140.49
11	1.93-2.11 (m, 2H)	1.93-2.11 (m, 2H)	28.30	28.31
12	0.96-1.20 (m, 2H)	0.96-1.20 (m, 2H)	22.31	22.31
13	0.96-1.20 (m, 2H)	0.96-1.20 (m, 2H)	40.12	40.12
14	--	--	45.19	45.19
15	0.48 (s, 3H)	0.48 (s, 3H)	11.73	11.72
16	2.78-2.82 (d, <i>J</i> = 16 Hz, H)	2.78-2.82 (d, <i>J</i> = 16 Hz, H)	55.92	55.92
17, 18, 22, 23, 24	1.25-1.46 (m, 10H)	1.25-1.46 (m, 10H)	23.03, 27.37, 35.60, 23.24, 39.91	23.03, 27.37, 35.60, 23.24, 39.91
19, 20, 25	2.29-2.47 (m, 3H)	2.29-2.47 (m, 3H)	55.57, 35.52, 27.14	55.57, 35.52, 27.14
21	0.87 (d, <i>J</i> = 6 Hz, 3H)	0.87 (d, <i>J</i> = 6 Hz, 3H)	18.58	18.58
26, 26'	0.85 (m, <i>J</i> = 8 Hz, 6H)	0.85 (m, <i>J</i> = 8 Hz, 6H)	22.58, 21.77	22.58, 21.77
27(OH)	5.02 (s)	5.02 (s)	--	--

s- singlet, d-doublet, and m-multiplet.

## Conclusions

The Trivedi Effect®-Consciousness Energy Healing Treatment on cholecalciferol/vitamin D<sub>3</sub> by the renowned Biofield Energy Healer, Mr. Mahendra Kumar Trivedi showed the astonishing significant impact on the isotopic abundance ratios and relative peak intensities. The LC-ESI-MS analysis of both the samples showed the mass of protonated ion at  $m/z$  385.3 (calcd for C<sub>27</sub>H<sub>45</sub>O<sup>+</sup>, 385.35) at the retention time (R<sub>t</sub>) 20.7 minutes. The liquid chromatographic peak area of the Biofield Energy Treated sample was significantly improved by 16.00% compared to the control sample. Similarly, the isotopic abundance ratio of P<sub>M+1</sub>/P<sub>M</sub> (<sup>2</sup>H/<sup>1</sup>H or <sup>13</sup>C/<sup>12</sup>C or <sup>17</sup>O/<sup>16</sup>O) was very significantly increased by 470.88% in the Biofield Energy Treated sample compared to the control sample. But, the isotopic abundance ratio of P<sub>M+2</sub>/P<sub>M</sub> (<sup>18</sup>O/<sup>16</sup>O) in the Biofield Energy Treated sample also significantly decreased by 41.81% compared to the control sample. Therefore, the <sup>2</sup>H, <sup>13</sup>C, and <sup>17</sup>O contributions from C<sub>27</sub>H<sub>45</sub>O<sup>+</sup> to  $m/z$  386 was significantly increased, whereas <sup>18</sup>O contribution from C<sub>21</sub>H<sub>21</sub>O<sub>6</sub><sup>+</sup> to  $m/z$  387 was decreased in the Biofield Energy Treated sample compared with the control sample. The GC-MS analysis showed that the parent molecular mass peak intensities ( $m/z$  384.4) in the Biofield Energy Treated sample at R<sub>t</sub> 23.04 and 23.65 minutes were significantly increased by 6.11% and 9.62%, respectively compared with the control sample. The proton and carbon signals for CH<sub>3</sub>, CH<sub>2</sub>, CH, C-OH, and =C= groups in the <sup>1</sup>H and <sup>13</sup>C NMR spectra of the control and Biofield Energy Treated samples were similar. The altered isotopic abundance ratios and mass peak intensities might be due to energy produced by The Trivedi Effect®-Consciousness Energy Healing Treatment *via* the possible mediation of neutrinos, which further lead to the change in the kinetic isotope effects and physicochemical properties of the Biofield Energy Treated sample. Thus, The Trivedi Effect®-Consciousness Energy Healing Treated vitamin D<sub>3</sub> would be advantageous for designing better nutraceutical and pharmaceutical formulations which might provide better therapeutic response against vitamin D deficiency, rickets, osteoporosis, arthritis, diabetes mellitus, cancer, cardiovascular diseases, mental disorders, inflammations, infections, stress, aging, glucose intolerance, multiple sclerosis, Parkinson's and Alzheimer's diseases, dementia, cognitive impairment in older adults, etc.

**Acknowledgements:** The authors are grateful to GVK Biosciences Pvt. Ltd., Trivedi Science, Trivedi Global, Inc., Trivedi Testimonials, and Trivedi Master Wellness for their assistance and support during this work.

## References

- Marx, J., Walls, R., Hockberger, R. Rosen's Emergency Medicine-Concepts and Clinical Practice, Volume 2, 8<sup>th</sup>Edn. (2013) Elsevier Health Sciences, US.  
PubMed | CrossRef | Others
- "WHO Model List of Essential Medicines" (2019) World Health Organization 205: 126.  
PubMed | CrossRef | Others
- Rosenkranz, H.S., Carr, H.S. Silver Sulfadiazine: Effect on the Growth and Metabolism of Bacteria. (1972) Antimicrob Agents Chemother 2(5): 367-372.  
PubMed | CrossRef | Others
- Wysor, M.S., Zollinhofer, R.E. On the mode of action of silver sulfadiazine. (1972) Pathobiol 38: 296-308.  
PubMed | CrossRef | Others
- "Silver Sulfadiazine". Drugs.com. Retrieved 15 May 2018.  
PubMed | CrossRef | Others
- [https://en.wikipedia.org/wiki/Silver\\_sulfadiazine](https://en.wikipedia.org/wiki/Silver_sulfadiazine).
- Charles, H.N., James, E.A., Milo, G. Physiologic surface active agents and drug absorption VIII: Effect of bile flow on sulfadiazine absorption in the rat. (1971) J Pharm Sci 60(1): 145-147.  
PubMed | CrossRef | Others
- Trivedi, M.K., Branton, A., Trivedi, D., et al. Evaluation of Physicochemical, Thermal, Structural, and Behavioral Properties of Magnesium Gluconate Treated with Energy of Consciousness (The Trivedi Effect®). (2017) J Drug Design Med Chem 3: 5-17.  
PubMed | CrossRef | Others
- Trivedi, M.K., Branton, A., Trivedi, D., et al. A comprehensive physical, spectroscopic, and thermal characterization of *Withania somnifera* (ashwagandha) root extract treated with the energy of consciousness (The Trivedi Effect®). (2017) Int J Biomed Mater Res 5: 5-14.  
PubMed | CrossRef | Others
- Trivedi, M.K., Branton, A., Trivedi, D., et al. Evaluation of physicochemical, spectral, thermal and behavioral properties of the biofield energy healing treated sodium selenate. (2017) Sci J Chem 5(2): 12-22.  
PubMed | CrossRef | Others
- Trivedi, M.K., Patil, S., Shettigar, H., et al. Spectroscopic characterization of chloramphenicol and tetracycline: An impact of biofield. (2015) Pharm Anal Acta 6: 395.  
PubMed | CrossRef | Others
- Trivedi, M.K., Mohan, T.R.R. Biofield energy signals, energy transmission and neutrinos. (2016) Am J Modern Phys 5(6): 172-176.  
PubMed | CrossRef | Others
- Rubik, B. The biofield hypothesis: Its biophysical basis and role in medicine. (2002) J Altern Complement Med 8: 703-717.  
PubMed | CrossRef | Others
- Nemeth, L. Energy and biofield therapies in practice. (2008) Beginnings 28: 4-5.  
PubMed | CrossRef | Others
- Rubik, B., Muehsam, D., Hammerschlag, R., et al. Biofield science and healing: history, terminology, and concepts. (2015) Glob Adv Health Med 4(Suppl): 8-14.  
PubMed | CrossRef | Others
- Koithan, M. Introducing complementary and alternative therapies. (2009) J Nurse Pract 5: 18-20.  
PubMed | CrossRef | Others
- Barnes, P.M., Bloom, B., Nahin, R.L. Complementary and alternative medicine use among adults and children: United States, 2007. (2008) Natl Health Stat Report 12: 1-23.  
PubMed | CrossRef | Others
- Trivedi, M.K., Branton, A., Trivedi, D., et al. Isotopic abundance ratio analysis of 1,2,3-trimethoxybenzene

- (TMB) after biofield energy treatment (The Trivedi Effect®) using gas chromatography-mass spectrometry. (2016) Am J Appl Chem 4(4): 132-140.  
PubMed | [CrossRef](#) | [Others](#)
19. Trivedi, M.K., Tallapragada, R.M., Branton, A., et al. Physicochemical and atomic characterization of silver powder after biofield treatment. (2015) J Bioengineer Biomedical Sci 5: 165.  
PubMed | [CrossRef](#) | [Others](#)
20. Trivedi, M.K., Nayak, G., Patil, S., et al. Studies of the atomic and crystalline characteristics of ceramic oxide nano powders after bio field treatment. (2015) Ind Eng Manage 4: 161.  
PubMed | [CrossRef](#) | [Others](#)
21. Trivedi, M.K., Branton, A., Trivedi, D., et al. Antibiofilm, biochemical reactions and genotyping characterization of biofield treated *Staphylococcus aureus*. (2015) Am J Bio Sci 3: 212-220.  
PubMed | [CrossRef](#) | [Others](#)
22. Trivedi, M.K., Branton, A., Trivedi, D., et al. Antimicrobial sensitivity, biochemical characteristics and biotyping of *Staphylococcus saprophyticus*: An impact of biofield energy treatment. (2015) J Women's Health Care 4: 271.  
PubMed | [CrossRef](#) | [Others](#)
23. Trivedi, M.K., Branton, A., Trivedi, D., et al. Agronomic characteristics, growth analysis, and yield response of biofield treated mustard, cowpea, horse gram, and groundnuts. (2015) Intl J Genet Genom 3: 74-80.  
PubMed | [CrossRef](#) | [Others](#)
24. Trivedi, M.K., Branton, A., Trivedi, D., et al. Evaluation of plant growth, yield and yield attributes of biofield energy treated Mustard (*Brassica juncea*) and Chick pea (*Cicer Arietinum*) Seeds. (2015) Agricult Forest Fisheries 4: 291-295.  
PubMed | [CrossRef](#) | [Others](#)
25. Branton, A., Jana, S. The use of novel and unique biofield energy healing treatment for the improvement of poorly bioavailable compound, berberine in male Sprague Dawley rats. (2017) Am J Clin Exper Med 5(4): 138-144.  
PubMed | [CrossRef](#) | [Others](#)
26. Branton, A., Jana, S. The influence of energy of consciousness healing treatment on low bioavailable resveratrol in male Sprague Dawley rats. (2017) Intl J Clin Develop Anatomy 3(3): 9-15.  
PubMed | [CrossRef](#) | [Others](#)
27. Trivedi, M.K., Branton, A., Trivedi, D., et al. Determination of isotopic abundance ratio of biofield energy treated 1,4-dichlorobenzene using gas chromatography-mass spectrometry (GC-MS). (2016) Modern Chem 4: 30-37.  
PubMed | [CrossRef](#) | [Others](#)
28. Trivedi, M.K., Branton, A., Trivedi, D., et al. Gas chromatography-mass spectrometric analysis of isotopic abundance of <sup>13</sup>C, <sup>2</sup>H, and <sup>18</sup>O in biofield energy treated *p*-tertiary butylphenol (PTBP). (2016) Am J Chem Engg 4: 78-86.  
PubMed | [CrossRef](#) | [Others](#)
29. Schellekens, R.C., Stellaard, F., Woerdenbag, H.J., et al. Applications of stable isotopes in clinical pharmacology. (2011) Br J Clin Pharmacol 72: 879-897.  
[PubMed](#) | [CrossRef](#) | [Others](#)
30. Weisel, C.P., Park, S., Pyo, H., et al. Use of stable isotopically labeled benzene to evaluate environmental exposures. (2003) J Expo Anal Environ Epidemiol 13(5): 393-402.  
[PubMed](#) | [CrossRef](#) | [Others](#)
31. Muccio, Z., Jackson, G.P. Isotope ratio mass spectrometry. (2009) Analyst 134(2): 213-222.  
[PubMed](#) | [CrossRef](#) | [Others](#)
32. Rosman, K.J.R., Taylor, P.D.P. Isotopic compositions of the elements 1997 (Technical Report). (1998) Pure Appl Chem 70: 217-235.  
[PubMed](#) | [CrossRef](#) | [Others](#)
33. Smith, R.M. Understanding Mass Spectra: A Basic Approach, Second Edition. (2004) John Wiley & Sons, Inc.  
[PubMed](#) | [CrossRef](#) | [Others](#)
34. Jürgen, H. Gross Mass Spectrometry: A Textbook (2<sup>nd</sup>Edn). (2004) Springer: Berlin.  
[PubMed](#) | [CrossRef](#) | [Others](#)
35. Pfeifer, T., Tuerk, J., Bester, K., et al. Determination of selected sulfonamide antibiotics and trimethoprim in manure by electrospray and atmospheric pressure chemical ionization tandem mass spectrometry. (2002) Rapid Commun Mass Spectrom 16(7): 663-669.  
[PubMed](#) | [CrossRef](#) | [Others](#)

Submit your manuscript to Omega Publishers and we will help you at every step:

- We accept pre-submission inquiries
- Our selector tool helps you to find the most relevant journal
- We provide round the clock customer support
- Convenient online submission
- Thorough peer review
- Inclusion in all major indexing services
- Maximum visibility for your research

Submit your manuscript at



<https://www.omegaonline.org/submit-manuscript>