Structural Characterization and Isotopic Abundance Ratios Analysis of Nanocurcumin Treated With Consciousness Energy Healing Treatment

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Abstract

Nanocurcumin possesses diverse pharmacological effects, including anti-inflammatory, antioxidant, antiangiogenic activities, etc. The objective of the experiment was to investigate the impact of the Trivedi Effect-Consciousness Energy Healing Treatment on the isotopic abundance ratios along with the structural properties of nanocurcumin using sophisticated spectroscopy techniques. Nanocurcumin powder was divided into two parts—one part of the sample was termed as control, while the other part was received the Trivedi Effect-Consciousness Energy Healing Treatment by a famous Biofield Energy Healer, Mr. Mahendra Kumar Trivedi and termed as treated sample. The LC-ESI-MS analysis of both the control and treated nanocurcumin exhibited the protonated molecular ion mass at m/z 369 (calcd for C₉H₆O₃, 369.13) at retention time (R) 18.98 minutes along with similar fragmentation pattern. However, the relative peak intensities of the fragmented ion peaks of the treated nanocurcumin were significantly altered compared to the control sample. The isotopic abundance ratio of P₁/P₀ (1₂H/₁₁C) was significantly increased by 76.54% in the treated nanocurcumin compared to the control sample. Therefore, the ₁₂C, ₁₆O, and ₁₈O contributions from C₉H₆O₃ to m/z 370 in the treated sample was significantly increased compared to the control sample. On the contrary, the isotopic abundance ratio of P₁/P₀ (₁₈O₁₂O) in the treated sample was significantly decreased by 28.39% compared to the control sample. Therefore, the ₁₈O contribution from C₉H₆O₃ to m/z 371 in the treated nanocurcumin was significantly decreased compared with the control sample. The GC-MS analysis showed that the chromatographic peak area% was significantly increased by 31.49% in the treated nanocurcumin compared to the control sample. The isotopic abundance ratio of P₁/P₀ (₁₂H₁₈O) in the treated sample was significantly increased by 37.60% compared to the control sample. Therefore, the ₁₂H contributions from C₉H₆O₃ to m/z 373 in the treated nanocurcumin was significantly decreased compared with the control sample. The isotopic abundance ratio of P₁/P₀ (₁₂H₁₆O) in the treated sample was significantly increased by 31.49% compared to the control sample. Therefore, the ₁₂H contributions from C₉H₆O₃ to m/z 374 in the treated nanocurcumin was significantly decreased compared with the control sample. The isotopic abundance ratio of P₁/P₀ (₁₂H₁₈O) in the treated sample was significantly increased by 37.60% compared to the control sample. Therefore, the ₁₂H contributions from C₉H₆O₃ to m/z 375 in the treated nanocurcumin was significantly decreased compared with the control sample.

Introduction

Curcumin (diferuloylmethane) is a polyphenolic compound isolated from the dietary spice turmeric, i.e., rhizomes of Turmeric (Curcuma longa Linn) [1]. Curcuma longa rhizome contains linear diarylethenoid, with molecules such as curcumin or derivatives of curcumin with different chemical groups called curcuminoids (2-6% of the rhizome). Curcuminoids contain 77% curcumin, 17% demethoxycurcumin, and 6% bisdemethoxycurcumin (Figure 1) [2,3]. Curcumin exhibits two tautomeric forms having a predominant keto form in neutral and acidic solutions, and stable enol form in alkaline medium [2]. Curcumin is the principal bioactive metabolite of Curcuma longa. From the ancient age, turmeric has been used for many ailments, particularly as an anti-inflammatory agent [2]. Curcumin tested as a potent antioxidant [4], besides it also shows the anti-inflammatory [5], antirheumatic [6], antimicrobial, antiangiogenic [1], and antitumor [7] activities. Furthermore, curcumin is also well established for the hepato- and nephro-protective [5], myocardial infarction protective [9], thrombosis suppressing [10], and hypoglycemia [11].

In spite of broad the therapeutic effectiveness and promising therapeutic index, the curcumin is limited due to its poor absorption from the gastrointestinal tract (GIT) and poor bioavailability due to its rapid metabolism in the liver and intestinal wall [1,2]. Several kinds of the literature indicated that curcumin administered orally absorbed from the GIT, and present in the general blood circulation after being metabolized to the form of glucuronide and glucuronide/sulfate conjugates.

[1,2,12,13]. Nanocurcumin is a modified form of curcumin where the particles of curcumin are transformed into nanoparticles that are practically more soluble and deliverable in the body. These curcumin nanoparticles have been shown to be more targeted to the tissue of interest that leads to better drug delivery and faster treatment without any wastage or side effects. To improve the bioavailability of curcumin, numerous approaches have been undertaken. Numerous published literature indicated that the Trivedi Effect-Consciousness Energy Healing Treatment has the impact on the various properties of drug substances that would be helpful in the modification of physicochemical, spectral, and thermal properties of pharmaceutical/nutraceutical compounds [14-17]. Biofield Energy is a unique para-dimensional electromagnetic field exists in the human body, resulting in the continuous emission of energy from the body, which can freely flow between the human and environment [18,19]. Different religions, have recognized a living force that preserves and inhabits every living organism and believed to co-relate with the soul, spirit, and mind since from the ancient times. Scientifically, this hypothetical vital living force has been evaluated and is considered as Bioenergetics Field. There are several types of Biofield Energy Healing Therapies that are known for their significant positive impacts on various disease [20]. The Biofield Energy Healers have the ability to harness the energy from the "Universal Energy Field" and can transmit this energy into any living or non-living object(s), which respond to meaningful way, and the process is called Biofield Energy Healing Treatment. Such type of energy therapies are recommended by the National Institute of Health/National Center for Complementary and Alternative Medicine (NIH/NCCAM), and they included them under the Complementary and Alternative Medicine (CAM) due to their several advantages [21].

In recent years the Trivedi Effect-Biofield Energy Treatment have been scientifically reported with remarkable and outstanding results in the field of biotechnology [22,23], microbiology [24,25], pharmaceutical/nutraceutical sciences [14-16], medical science [26,27], materials science [28,29], agriculture [30,31], etc. The Trivedi Effect also found to have a remarkable effect of altering the isotopic abundance ratios of various compounds may be through the possible mediation of neutrinos [17,32,33]. Study on the natural stable isotope ratio analysis has wide applications in several fields of sciences to understand the isotope effects resulting from the alterations of the isotopic composition [34-36]. Gas chromatography-mass spectrometry (GC-MS) and liquid chromatography-mass spectrometry (LC-MS), are widely used for the analysis of isotope ratio with sufficient precision [35]. Therefore, in this experiment, the LC-MS based isotopic abundance ratio analysis of \(P_{\text{H}}/P_{\text{H}}^{12} (\text{H}/\text{H})\) or \(^{13}C/^{12}C\) or \(^{18}O/^{16}O\) and \(P_{\text{H}}/P_{\text{H}}^{12} (\text{H}/\text{H})\) samples was performed to evaluate the influence of the Trivedi Effect-Consciousness Energy Healing Treatment on the isotopic abundance ratio in nanocurcumin. Consequently, the LC-MS, GC-MS, and NMR (Nuclear Magnetic Resonance) techniques were also used to characterize the structural properties of the nanocurcumin.

Materials and Methods

Chemicals and Reagents

The test sample nanocurcumin (40%) powder was purchased from Sanat Products Ltd., India, and other chemicals also used in the experiment were of analytical grade purchased in India.

Consciousness Energy Healing Treatment Strategies

The test sample nanocurcumin was equally divided into two parts. One part of nanocurcumin was termed as Biofield Energy Treated sample, which received the Consciousness Energy Healing Treatment by a famous Biofield Energy Healer, Mr. Mahendra Kumar Trivedi (USA) remotely under the standard laboratory conditions for 3 minutes. Besides, the other part of nanocurcumin was not received the Trivedi Effect-Consciousness Energy Healing Treatment denoted as the control sample. But it was treated by a "sham" healer under the similar laboratory conditions, who did not have any knowledge about the Biofield Energy. Consequently, both the nanocurcumin samples were kept in similar sealed conditions and further analyzed by using sophisticated analytical techniques.

Characterization

Liquid Chromatography-Mass Spectrometry (LC-MS) Analysis and Calculation of Isotopic Abundance Ratio

The LC-MS analysis of both the nanocurcumin samples was carried out with the help of LC-Dionex Ultimate 3000, MS-TSQ Endura (USA) having a photo-diode array (PDA) detector connected with a Thermo Scientific TSQ Endura (USA) triple-stage quadrupole mass spectrometer with heated electrospray ionization (ESI) probe. The separation made in a reversed-phase Zorbax SB-C18 100 × 4.6mm × 3.5μm column (40°C). 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 injection volume was 10μL, and the total run time was 35 min. Chromatographic separation was achieved using gradient condition as follow: 0 min-5% B, 5 min-5% B, 15 min-60% B, 20 min-95% B, 25 min-95% B, 30 min-5% B, and 35 min-5% B. The mass spectrometric analysis was performed under +ve ESI mode.

The natural abundance of the common elements C, O, and H are obtained from the literature [37-40]. The isotopic abundance ratios \((P_{\text{H}}/P_{\text{H}}^{12} / P_{\text{H}}^{13} / P_{\text{H}}^{12})\) for the control and Biofield Energy Treated nanocurcumin was calculated.

\[
\% \text{change in isotopic abundance ratio} = \left[ \frac{\text{IAR}_{\text{treated}} - \text{IAR}_{\text{control}}}{\text{IAR}_{\text{control}}} \right] \times 100
\]

Figure 1: Curcuminoids from the rhizomes of Curcuma longa.
Gas Chromatography-Mass Spectrometry (GC-MS) Analysis

An Agilent 7890B Gas chromatograph equipped with a silica capillary column (30 m x 0.25mm) (HP-5 MS) and coupled to a quadrupole detector with pre-filter (5977B, USA) was operated with electron impact (EI) ionization in positive/negative mode at 70 eV was used for the analysis. The oven temperature was programmed from 50°C (1 min hold) to 300°C @ 20°C /min to 200°C (10 minutes hold). Temperatures of the injector, detector (FID), auxiliary, ion source, and quadrupole detector were 250,300,280,230, and 150°C.

Nanocurcumin was dissolved in methanol, and 1.0µL was splitlessly injected with helium as a carrier gas with a flow rate of 1.5 mL/min.

Nuclear Magnetic Resonance (NMR) Analysis

$^1$H NMR spectra of nanocurcumin were recorded at 400 MHz on Agilent-MRDD2 FT-NMR. Chemical shifts (δ) were in parts per million (ppm) relative to the solvent’s residual proton chemical shift {(CD$_3$)$_2$SO, δ = 2.5}. Similarly, $^{13}$C NMR spectra of nanocurcumin were measured at 100 MHz on Agilent-MRDD2 FT-NMR spectrometer at room temperature. Chemical shifts (δ) were in parts per million (ppm) relative to the solvent’s residual carbon chemical shift {(CD$_3$)$_2$SO, δ = 39.52}.

Results and Discussion

Liquid Chromatography-Mass Spectrometry (LC-MS) Analysis

The LC-MS chromatograms of both the nanocurcumin samples showed three sharp peaks (Figure 2) at the retention times (R$_t$) of 18.51, 18.74, and 18.98 minutes. The %peak area at R$_t$ 18.51, 18.74, and 18.98 minutes was 4.56, 25.68, and 68.44 in control and 4.21, 25.90, 68.52 in the Biofield Energy Treated sample, respectively. It indicated that the polarity of the Biofield Energy Treated sample remained close

<table>
<thead>
<tr>
<th>Parameter</th>
<th>Control sample</th>
<th>Biofield Energy Treated sample</th>
<th>% Change</th>
</tr>
</thead>
<tbody>
<tr>
<td>PM at m/z 369 (%)</td>
<td>100</td>
<td>100</td>
<td>0.00</td>
</tr>
<tr>
<td>PM+1 at m/z 370 (%)</td>
<td>19.95</td>
<td>35.22</td>
<td>76.54</td>
</tr>
<tr>
<td>PM+1/PM</td>
<td>0.1995</td>
<td>0.3522</td>
<td>76.54</td>
</tr>
<tr>
<td>PM+2 at m/z 371 (%)</td>
<td>6.41</td>
<td>4.59</td>
<td>-28.39</td>
</tr>
<tr>
<td>PM+2/PM</td>
<td>0.0641</td>
<td>0.0459</td>
<td>-28.39</td>
</tr>
</tbody>
</table>

PM = the relative peak intensity of the parent molecular ion M$^+$; PM+1 = the relative peak intensity of the isotopic molecular ion [M+1]$^+$; PM+2 = the relative peak intensity of the isotopic molecular ion [M+2]$^+$, and M = mass of the parent curcumin molecule.

Table 2: GC-MS chromatographic and mass spectra analysis at R$_t$, 10.68 minutes of the control and treated

<table>
<thead>
<tr>
<th>Parameters</th>
<th>Control sample</th>
<th>Biofield Energy Treated sample</th>
<th>% Change</th>
</tr>
</thead>
<tbody>
<tr>
<td>Peak area%</td>
<td>18.83</td>
<td>24.76</td>
<td>31.49</td>
</tr>
<tr>
<td>Mass peak intensity at m/z 177</td>
<td>40502.12</td>
<td>55730.65</td>
<td>37.60</td>
</tr>
<tr>
<td>Mass peak intensity at m/z 192</td>
<td>43361.76</td>
<td>61938.27</td>
<td>42.84</td>
</tr>
</tbody>
</table>

Table 3: $^1$H and $^{13}$C NMR spectroscopic data of the control and treated nanocurcumin.

<table>
<thead>
<tr>
<th>Position</th>
<th>$^1$H NMR δ (ppm)</th>
<th>$^{13}$C NMR δ (ppm)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Control</td>
<td>Treated</td>
<td>Control</td>
</tr>
<tr>
<td>1, 1’</td>
<td>br, s(9.64)</td>
<td>br, s(9.64)</td>
</tr>
<tr>
<td>2, 2’</td>
<td>s(3.84)</td>
<td>s(3.84)</td>
</tr>
<tr>
<td>3, 3’</td>
<td>--</td>
<td>--</td>
</tr>
<tr>
<td>4, 4’</td>
<td>--</td>
<td>--</td>
</tr>
<tr>
<td>5, 5’</td>
<td>s(7.32)</td>
<td>s(7.32)</td>
</tr>
<tr>
<td>6, 6’</td>
<td>--</td>
<td>--</td>
</tr>
<tr>
<td>7, 7’</td>
<td>d(7.15, J = 8)</td>
<td>d(7.15, J = 8)</td>
</tr>
<tr>
<td>8, 8’</td>
<td>d(6.82, J = 8)</td>
<td>d(6.82, J = 8)</td>
</tr>
<tr>
<td>9, 9’</td>
<td>d(7.54, J = 16)</td>
<td>d(7.55, J = 24)</td>
</tr>
<tr>
<td>10, 10’</td>
<td>d(6.72, J = 8)</td>
<td>d(6.72, J = 8)</td>
</tr>
<tr>
<td>11, 11’</td>
<td>--</td>
<td>--</td>
</tr>
<tr>
<td>12</td>
<td>s(6.05)</td>
<td>s(6.06)</td>
</tr>
</tbody>
</table>

Where IAR = isotopic abundance ratio in control and treated nanocurcumin.

Gas Chromatography-Mass Spectrometry (GC-MS) Analysis

An Agilent 7890B Gas chromatograph equipped with a silica capillary column (30 m x 0.25mm) (HP-5 MS) and coupled to a quadrupole detector with pre-filter (5977B, USA) was operated with electron impact (EI) ionization in positive/negative mode at 70 eV was used for the analysis. The oven temperature was programmed from 50°C (1 min hold) to 300°C @ 20°C /min to 200°C (10 minutes hold). Temperatures of the injector, detector (FID), auxiliary, ion source, and quadrupole detector were 250,300,280,230, and 150°C.
compared with the control nanocurcumin.

The ESI-MS spectra of the control and Biofield Energy Treated samples at R_t of 18.51 minutes exhibited the presence of the molecular mass of bisdemethoxy curcumin adduct with hydrogen ion (Figure 3) at m/z 309 (calcld for C_{19}H_{16}O_{4}+, 309.11). Similarly, at R_t of 18.74 minutes showed the presence of the molecular mass of demethoxy curcumin adduct with hydrogen ion (Figure 3) at m/z 339 (calcld for C_{20}H_{19}O_{5}+, 339.12). Curcuminoids are the molecules such as curcumin or derivatives of curcumin with different chemical groups that have been formed to increase the solubility of curcumin and make them suitable for drug formulation [1,2].

At the R_t of 18.98 minutes the corresponding molecular mass peak at m/z 369 (calcld for C_{21}H_{21}O_{6}+, 369.13) was found to be curcumin adduct with hydrogen ion (Figures 4 and 5). Other lower mass peak at m/z 245 (calcld for C_{14}H_{13}O_{4}+, 245.08 or C_{19}H_{17}+, 245.08) and 177 (calcld for C_{10}H_{9}O_{3}+, 177.05) in the control and Biofield Energy Treated samples were observed (Figures 4 and 5).

The ESI-MS spectra of the control and Biofield Energy Treated nanocurcumin showed almost similar type of mass fragmentation pattern (Figures 4 and 5). The molecular ion peak at m/z 369 exhibited 100% relative base peak intensity in both ESI-MS spectra (Figure 4). The relative peak intensities of the other ion peaks in the Trivedi Effect®-Consciousness Energy Healing Treated nanocurcumin were significantly altered compared to the control sample.

**Isotopic Abundance Ratio Analysis**

The control and Biofield Energy Treated samples of nanocurcumin showed the mass of a protonated molecular ion at m/z 369 (C_{21}H_{21}O_{6}+) with 100% relative abundance in the mass spectra. The theoretical calculation of isotopic peak P_{M+1} for the protonated nanocurcumin presented as below:

\[
P(^1\text{C}) = \left[\left(21 \times 1.1\%\right) \times 100\%\right] / 100\% = 23.1\%
\]

\[
P(^2\text{H}) = \left[\left(21 \times 0.015\%\right) \times 100\%\right] / 100\% = 0.315\%
\]

\[
P(^17\text{O}) = \left[\left(6 \times 0.04\%\right) \times 100\%\right] / 100\% = 0.24\%
\]
P_{M+1} i.e.,^{13}\text{C}, ^{2}\text{H}, and ^{17}\text{O} contributions from C_{21}\text{H}_{21}\text{O}_{6}^{+} to m/z 370 = 23.66%.

Similarly, the theoretical calculation of isotopic peak P_{M+2} for the protonated nanocurcumin presented as below:

\[
P^{(18}\text{O}) = [(6 \times 0.20\%) \times 100\%] / 100\% = 1.2\%
\]

\[
P_{M+2} \text{ of}^{18}\text{O} \text{contribution from C}_{21}\text{H}_{21}\text{O}_{6}^{+} \text{to} \text{m/z} 371 = 1.2\%
\]

The calculated isotopic abundance of P_{M+1} value 23.66% was closer to the observed value (19.95%), but the calculated P_{M+2} value 1.2% was lower to the observed value (6.41%) (Table 1). Thus, 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., ^{13}\text{C}, ^{2}\text{H}, ^{18}\text{O}, and ^{17}\text{O}) contributions to the mass of the isotopic molecular ion [M+1]⁺ and [M+2]⁺. ^{2}\text{H} did not contribute much any isotopic m/z ratios because of its less natural abundance compared to the natural abundances of C and O isotopes [38,40]. From the calculations, it is confirmed that ^{13}\text{C}, ^{17}\text{O}, and ^{18}\text{O} have the major contributions from nanocurcumin to the isotopic mass peak at m/z 370 and 371. Therefore, P_{M+1}, P_{M+1}^{(13}\text{C}), P_{M+1}^{(17}\text{O)}, and P_{M+1}^{(18}\text{O}) of the nanocurcumin at m/z 369, 370, and 371 of the control and Biofield Energy Treated nanocurcumin were obtained from the experimental relative abundance of [M⁺], [(M+1)⁺], and [M+2]⁺ peaks, respectively in the mass spectra (Table 1).

The isotopic abundance ratio of P_{M+1}/P_{M} (^{13}\text{C}/^{12}\text{C} or ^{17}\text{O}/^{16}\text{O}) in the Trivedi Effect®-Consciousness Energy Healing Treatment nanocurcumin was significantly increased by 76.54% compared to the control sample (Table 1). Thus, the ^{13}\text{C}, ^{2}\text{H}, and ^{18}\text{O} contributions from C_{21}\text{H}_{21}\text{O}_{6}^{+} to m/z 370 in the Biofield Energy Treated sample was significantly increased compared to the control sample. On the contrary, the isotopic abundance ratio of P_{M+2}/P_{M} (^{18}\text{O}/^{16}\text{O}) in the Biofield Energy Treated nanocurcumin was significantly decreased by 28.39% compared to the control sample (Table 1). Therefore, the ^{18}\text{O} contribution from C_{21}\text{H}_{21}\text{O}_{6}^{+} to m/z 371 in the Biofield Energy Treated nanocurcumin was significantly decreased compared with the control sample. Alteration in the number of neutrons in the molecule leads to the increased or decreased isotopic abundance of the compounds. Thus, it can be assumed that due to the possible mediation of neutrinos oscillation, the atomic mass and the atomic charge alter. Therefore, it is expected that the Trivedi Effect®-Consciousness Energy Healing Treatment might provide the necessary energy for the neutrino oscillations leads to the modification of the fundamental physicochemical properties of a compound [17,32,33]. The change in the kinetic isotope effects leads to the alteration in the isotopic abundance ratio of the atoms/molecules, 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 [32,33,35]. The treated nanocurcumin with altered isotopic abundance ratio (P_{M+1}/P_{M} and P_{M+2}/P_{M}) might be advantageous for the better nutraceutical/pharmaceutical formulations.

**Gas Chromatography-Mass Spectrometry (GC-MS) Analysis**

The chromatograms of both the control and Biofield Energy

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**Figure 7:** GC-MS spectra of the control and treated nanocurcumin at Rt 10.68 minutes.

**Figure 8:** The 1H NMR spectra of the control and treated nanocurcumin.

**Figure 9:** The 13C NMR spectra of the control and treated nanocurcumin.
Treated nanocurcumin showed several peaks (Figure 6) with very close retention time indicated that the polarity of the Biofield Energy Treated sample remained same compared with the control nanocurcumin. The mass spectra of both the control and Biofield Energy Treated samples (Figure 7) showed that m/z 177 (calcd for C_{66}H_{47}O_{14}, 177.05) and 192 (calcd for C_{61}H_{45}O_{13}, 192.08) are fragments of nanocurcumin, at R$_t$ 10.68 minutes. The peak area% and mass peak intensities in the chromatograms and mass spectra, respectively of the Biofield Energy Treated nanocurcumin was significantly altered compared to the control sample (Table 2).

The chromatographic peak area% of the control and Biofield Energy Treated nanocurcumin was 18.83 and 24.76, respectively (Table 2) at R$_t$ 10.68 minutes. Therefore, the change in the peak area% was significantly increased by 31.49% in the Biofield Energy Treated nanocurcumin compared to the control sample. This indicated that the relative concentration of the nanocurcumin significantly increased compared to the control sample. The Trivedi Effect-Consciousness Energy Healing Treatment might have influenced the physicochemical properties of nanocurcumin leads to an increase in the solubility and concentration in the solution. Increase in the solubility and concentration may improve the bioavailability of the nanocurcumin with desired (anticipated) pharmacological response [41].

The mass peak intensities of some major fragmented mass peaks were significantly altered (Table 2) at m/z 177 and 192. The mass peak intensity of the Biofield Energy Treated sample at m/z 177 (calcd for C_{66}H_{47}O_{14}, 177.05) was significantly increased by 37.60% compared to the control sample. Similarly, the mass peak intensity of the Biofield Energy Treated sample at m/z 192 (calcd for C_{61}H_{45}O_{13}, 192.08) was significantly increased by 42.84% compared to the control sample.

The mass peak intensities were significantly increased may be due to the impact of the Trivedi Effect-Consciousness Energy Healing Treatment. The Trivedi Effect is a scientifically proved phenomena which have the remarkable potential to alter the isotopic abundance ratios of various compounds might be through the possible mediation of neutrinos [17,32,33].

**Nuclear Magnetic Resonance (NMR) Spectroscopy Analysis**

The $^1$H and $^{13}$C NMR spectra are shown in Figure 8 and 9, respectively for the control and Biofield Energy Treated nanocurcumin. The $^1$H and $^{13}$C NMR analyzed spectral data of both the control and Biofield Energy Treated nanocurcumin are presented in Table 3. The $^1$H NMR spectra of the control and Biofield Energy Treated samples (Figure 8) indicated that signals for the protons coupling of CH$_2$, CH, OH, and OCH$_3$ protons of nanocurcumin were in the range of $\delta$ 3.84 to 9.64 ppm (Table 3), which were very close to each other. The experimental results were closely matched to the reported literature [42-44]. Similarly, the carbon signals for CH$_2$, CH, COH, OCH$_3$, and CO groups in the $^{13}$C NMR spectrum (Figure 9) of the Biofield Energy Treated sample were almost same compared to the control sample of nanocurcumin (Table 3). The $^1$H and $^{13}$C NMR results indicated that there was no structural modification of the Biofield Energy Treated nanocurcumin compared to the control sample.

**Conclusions**

The Trivedi Effect-Consciousness Energy Healing Treatment has shown the significant impact on nanocurcumin. The LC-ESI-MS analysis of both the control and Biofield Energy Treated nanocurcumin exhibited the protonated molecular ion mass at m/z 369 at R$_t$ 18.98 minutes along with similar fragmentation pattern. However, the relative peak intensities of the fragmented ion peaks of the Biofield Energy Treated nanocurcumin were significantly altered compared to the control sample. The isotopic abundance ratio of P$_{M+1}$/P$_M$ ($^{12}$C/$^{13}$C or $^{16}$O/$^{18}$O) was significantly increased by 76.54% in the Biofield Energy Treated nanocurcumin compared to the control sample. Therefore, the $^{12}$C, $^1$H, and $^{16}$O contributions from C$_{66}H_{47}O_{14}$ to m/z 370 in the Biofield Energy Treated sample was significantly increased compared to the control sample. On the contrary, the isotopic abundance ratio of P$_{M+1}$/P$_M$ ($^{18}$O/$^{16}$O) in the Biofield Energy Treated sample was significantly decreased by 28.39% compared to the control sample. Therefore, the $^{18}$O contribution from C$_{61}H_{45}O_{13}$ to m/z 371 in the Biofield Energy Treated nanocurcumin was significantly decreased compared with the control sample. The GC-MS analysis showed that the chromatographic peak area% was significantly increased by 31.49% in the Biofield Energy Treated nanocurcumin compared to the control sample at R$_t$ 10.68 minutes. The fragmented mass peak intensity of the Biofield Energy Treated sample at m/z 177 (C$_{66}H_{47}O_{14}$) and 192 (C$_{61}H_{45}O_{13}$) were significantly increased by 37.60% and 42.84%, respectively compared to the control sample. From the results, it is concluded that the Trivedi Effect-Consciousness Energy Healing Treatment might provide the necessary energy for the neutrino oscillations in nanocurcumin leads to the improvement of the therapeutic profile. The Trivedi Effect-Consciousness Energy Healing Treatment could be an economical approach in the design of better nutraceutical and pharmaceutical formulations. Thus, the treated nanocurcumin with altered isotopic abundance ratio and improved solubility profile could provide better therapeutic response against inflammation, cancer, rheumatism, hypoglycemia, microbial infections, the hepato- and nephro-damage, myocardial infarction, thrombosis suppression, hypoglycemic condition, etc.

**Acknowledgements**

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**References**

Para el conocimiento científico y académico, este documento proporciona información sobre la caracterización estructural y la isotopidad de abundancia de SmCurcumina tratar con conciencia energética.


