

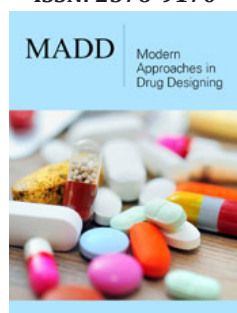
# Evaluation of the Impact of Consciousness Energy Healing Treatment on the Isotopic Abundance Ratio of Folic Acid Using LC-MS Spectrometry

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## Abstract

Folic acid (vitamin B<sub>9</sub> or folate) is one of the water-soluble B vitamins obtained from the foods and supplements, which have many functions in the body and also used for the prevention and treatment of folic acid deficiency disorders. This study was designed to investigate the impact of The Trivedi Effect®-Biofield Energy Healing Treatment on the structural properties and the isotopic abundance ratio of folic acid using LC-MS spectroscopy. The folic acid sample was divided into two parts, one part of folic acid was considered as the control sample (no Biofield Energy Treatment was provided), while the second part was treated with The Trivedi Effect®-Consciousness Energy Healing Treatment remotely by a renowned Biofield Energy Healer, Alice Branton and termed as the treated sample. The LC-MS spectra of both the samples at retention time (R<sub>t</sub>) 1.83 minutes exhibited the mass of the molecular ion peak at 441.92 (calcd for C<sub>19</sub>H<sub>20</sub>N<sub>7</sub>O<sub>6</sub><sup>+</sup>, 442.15) along with low molecular fragmented mass peaks at *m/z* 295.17, 267.17, 250.25, and 208 for C<sub>14</sub>H<sub>11</sub>N<sub>6</sub>O<sub>2</sub><sup>+</sup>, C<sub>12</sub>H<sub>15</sub>N<sub>2</sub>O<sub>5</sub><sup>+</sup>, C<sub>12</sub>H<sub>12</sub>NO<sub>3</sub><sup>+</sup>, and C<sub>11</sub>H<sub>14</sub>NO<sub>3</sub><sup>+</sup>, respectively were also observed. The isotopic abundance ratio of P<sub>M+1</sub>/PM (<sup>2</sup>H/<sup>1</sup>H or <sup>13</sup>C/<sup>12</sup>C or <sup>15</sup>N/<sup>14</sup>N or <sup>17</sup>O/<sup>16</sup>O) in the treated folic acid was significantly increased by 69.80% compared with the control sample.

Hence, the <sup>13</sup>C, <sup>2</sup>H, <sup>15</sup>N, and <sup>17</sup>O contributions from C<sub>19</sub>H<sub>20</sub>N<sub>7</sub>O<sub>6</sub><sup>+</sup> to *m/z* 442.75 in the Biofield Energy Treated sample were significantly increased compared with the control sample. The isotopic abundance ratio of P<sub>M+1</sub>/PM (<sup>2</sup>H/<sup>1</sup>H or <sup>13</sup>C/<sup>12</sup>C or <sup>15</sup>N/<sup>14</sup>N or <sup>17</sup>O/<sup>16</sup>O) in the Biofield Energy Treated folic acid was significantly increased compared to the control sample. It can be assumed that the changes in isotopic abundance and mass peak intensities due to changes in nuclei possibly through the interference of neutrino particles are *via* The Trivedi Effect® - Consciousness Energy Healing Treatment. The new form of Biofield Energy Treated folic acid would be better designing novel pharmaceutical formulations that might offer better therapeutic responses against glossitis, diarrhea, confusion, depression, anemia, gray hair, mouth sores, poor growth, swollen tongue, congenital deformities, embryonic defects, altered memory and brain function, leuco- and thrombocytopenia, cardiovascular disease, neural tube defects, and malignancies, risk of potentially developing allergic diseases, long-term risk of lower bone density, etc.

**Keywords:** Folic acid; The Trivedi Effect®, Biofield energy; Consciousness energy healing treatment; LC-MS

## Introduction

Folic acid (Vitamin B<sub>9</sub> or folate) is one of the water-soluble B vitamins obtained from the foods and supplements [1]. The daily amount of folate recommended for adults is 400 micrograms (mcg), while women in pregnancy advised to get 400 to 800mcg of folic acid a day [2]. The daily requirement of folate would be achieved using the food items containing the dark green leafy vegetables, fruits and fruit juices, nuts, soybeans, chickpeas, avocado, beetroot, spinach, liver, yeast, asparagus, kale, Brussels sprouts, dairy products, poultry and meat, eggs, seafood, grains, and some beers [3-6]. Folic acid acts as a cofactor are required for DNA replication, various intracellular reactions, and as a substrate for many enzymatic reactions included in vitamin metabolism and amino acid synthesis. Besides, folic acid is majorly involved in the synthesis and repair of genetic materials, i.e., DNA and RNA, aiding rapid cell division and growth, enhancing the overall brain health, and in age-related hearing loss [7]. In addition, folic acid plays an important role in normal blood formation, homocysteine levels, normal metabolism of the immune system, cell division, maintain normal maternal tissue growth during

pregnancy, normal amino acid synthesis, control various common psychological functions, reduction of tiredness and fatigue. Folic acid deficiency might lead to various common health problems and is one of the common vitamin deficiencies, which might result from inadequate intake, defective absorption (i.e., Crohn's disease or celiac disease), abnormal metabolism or increased requirements during pregnant or breastfeeding, some genetic disorders, certain medicines (i.e., phenytoin, sulfasalazine, or trimethoprim-sulfamethoxazole), alcohol consumption, etc. Its deficiency leads to glossitis, diarrhea, confusion, depression, anemia, gray hair, mouth sores, poor growth, swollen tongue, congenital deformities, embryonic defects, altered memory and brain function, leuco- and thrombocytopenia, cardiovascular disease, particular neural tube defects, and malignancies, higher risk of potentially developing allergic diseases, long-term risk of lower bone density, etc. [8-10]. The physicochemical properties of folic acid are very important to the nutraceutical and pharmaceutical formulations. The solubility profile of folic acid is very poor. It is almost insoluble in nature (1.6mg/L in water). Aqueous solutions of it are heat sensitive and decompose rapidly in the presence of light and /or riboflavin [11].

Improvement in the physicochemical properties of the pharmaceutical/ nutraceutical compounds are very much challenging and crucial factors. In this aspect, The Trivedi Effect®- Consciousness Energy Healing Treatment experimentally proved the improvement of bioavailability profile of many pharmaceutical/nutraceutical compounds, i.e., vitamin D<sub>3</sub>, resveratrol, and berberine in male Sprague Dawley rats [12-14]. The Trivedi Effect® is a natural and only scientifically proven phenomenon in which a person can harness this inherently intelligent energy and transmit it anywhere on the planet through the possible mediation of neutrinos [15]. Every living organism possesses a unique infinite, para-dimensional electromagnetic energy field surrounding the body known as Biofield Energy. The Biofield Energy Healers has the ability to harness the energy from the "Universal Energy Field" and can transmitted into any living or non-living object(s). Further, the object(s) respond to a useful way is known as the Biofield Energy Healing Treatment. There are several Biofield based Energy Healing Therapies that are used nowadays against various disease conditions [16-18]. 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, Qi Gong, Tai Chi, chiropractic/osteopathic manipulation, meditation, homeopathy, acupuncture, acupressure, healing touch, hypnotherapy, movement therapy, naturopathy, Reiki, cranial sacral therapy, etc. [19]. These therapies have been recognised by most of the U.S.A. population with several advantages [20]. The Trivedi Effect®- Energy of Consciousness Healing Treatment has the significant impact on the transformation, the characteristic properties of metals and ceramics [21,22], organic compounds [23,24], microorganisms [25,26], to improve the overall productivity of crops [27,28], and altered the isotopic abundance ratio [29,30].

To study the natural stable isotope ratio analysis has many applications in the different field of sciences to understand the isotope effects resulting from the alterations of the isotopic composition [31-33]. 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 [32]. The Trivedi Effect®-Biofield Energy Healing Treatment could be an economical approach for designing better pharmaceutical/nutraceutical formulations. Therefore, a LC-MS based structural characterization and the isotopic abundance ratio of  $P_{M+1}/P_M$  ( $^2\text{H}/^1\text{H}$  or  $^{13}\text{C}/^{12}\text{C}$  or  $^{17}\text{O}/^{16}\text{O}$ ) in The Trivedi Effect® - Consciousness Energy Healing Treated folic acid was performed in comparing its effect on a control sample.

## Materials and Methods

### Chemicals and reagents

Folic acid dihydrate (> 97%) was purchased from Alfa Aesar, India. All other chemicals used during the experiments were of analytical grade available in India.

### Consciousness energy healing treatment strategies

The folic acid dihydrate powder sample was the test sample divided into two parts. One part of folic acid powder sample was considered as a control sample (no Biofield Energy Treatment was provided). However, the other part of folic acid was exposed to The Trivedi Effect®-Consciousness Energy Healing Treatment remotely under standard laboratory conditions for 3 minutes and known as The Trivedi Effect® Treated (Biofield Energy Treated) folic acid. The Biofield Energy Treatment was provided through the healer's unique energy transmission process by the renowned Biofield Energy Healer, Alice Branton, USA, to the test sample. Further, the control sample was treated with "sham" healer for the comparison purpose. The sham healer did not have any knowledge about the Biofield Energy Treatment. After that, the Biofield Energy Treated and untreated folic acid samples were kept in sealed conditions and characterized using LC-MS analytical 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 folic acid was carried out with the help of LC-MS Thermo Fisher Scientific, USA equipped with an ion trap detector connected with a triple-stage quadrupole mass spectrometer. The column used here was a reversed phase Thermo Scientific Synchronis C18 (Length-250mm X ID 4.6mm X 5micron), maintained at 25°C. The diluent used for the sample preparation was methanol and water (1:1). The folic acid solution injection volume was 10µL and the analyte was eluted using acetonitrile +10mM ammonium acetate (85:15), pumped at a constant flow rate of 1 mL/min. Chromatographic separation was achieved using gradient condition and the total run time was 10 min. Peaks were monitored at 220nm using the PDA detector. Mass spectrometric analysis was performed under ESI +ve ion mode. The total

ion chromatogram, peak area % and mass spectrum of the individual peak which appeared in LC along with the full scan ( $m/z$  50-700) were recorded. The total ion chromatogram and mass spectrum of the individual peak (appeared in LC-MS) were recorded.

The natural abundance of each isotope (H, C, N, and O) can be predicted from the comparison of the height 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 [33-36]. The LC-MS based isotopic abundance ratio ( $P_{M+1}/P_M$ ) for the control and Biofield Energy Treated folic acid was calculated.

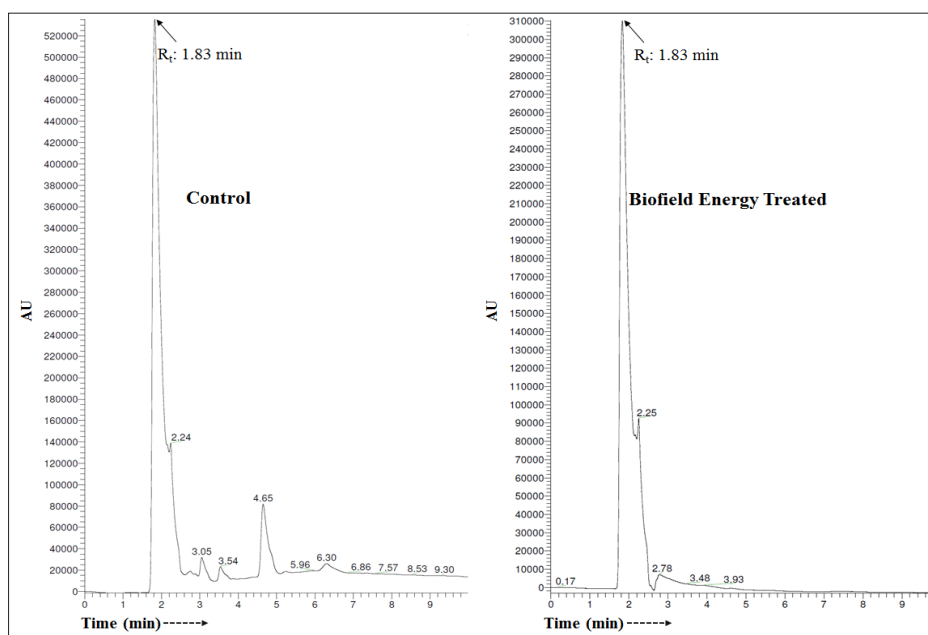
Percentage (%) change in isotopic abundance ratio =  $[(IAR_{Treated} - IAR_{Control}) / IAR_{Control}] \times 100$ ,

Where  $IAR_{Treated}$  = isotopic abundance ratio in the treated sample and  $IAR_{Control}$  = isotopic abundance ratio in the control sample.

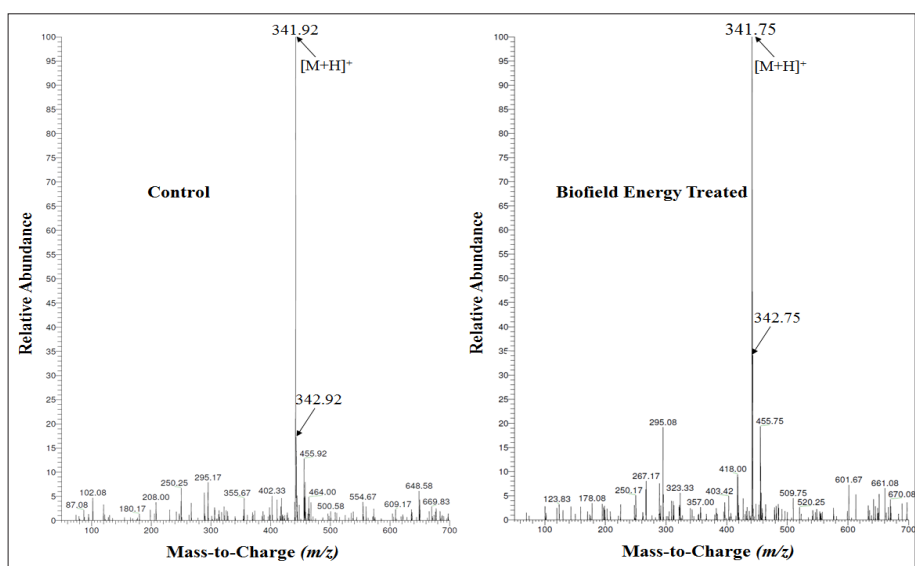
## Results and Discussion

### Liquid chromatography-mass spectrometry (LC-MS)

The LC-MS of the control and Biofield Energy Treated folic acid were carried out. The chromatograms of the control and Biofield Energy Treated folic acid showed the single major peak at retention time ( $R_t$ ) of 1.83 minutes in both the case (Figure 1). This result indicated that the polarity of both the samples was very close to each other.



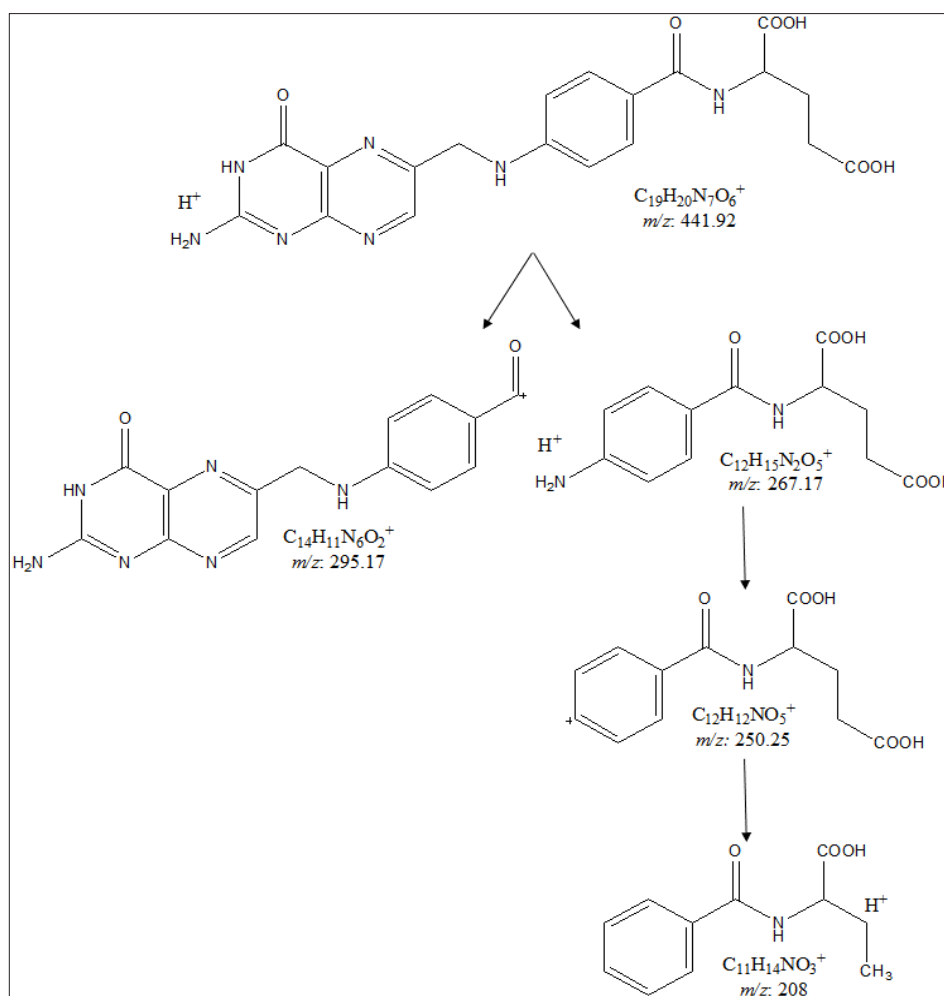
**Figure 1:** Liquid chromatograms of the control and Biofield Energy Treated folic acid.



**Figure 2:** Mass spectra of the control and Biofield Energy Treated folic acid at  $R_t$  1.83 minutes.

The mass spectra of both the samples of folic acid are shown in Figure 2. The mass spectra of the control and Biofield Energy Treated samples at  $R_t$  of 1.83 minutes exhibited the presence of the molecular ion of folic acid ( $C_{19}H_{20}N_7O_6^+$ ) adduct with hydrogen ion (Figure 2) at  $m/z$  441.92 (calcd for  $C_{19}H_{20}N_7O_6^+$ , 442.15). Along with

the molecular mass peak, low molecular fragmented mass peaks at  $m/z$  295.17, 267.17, 250.25, and 208 for  $C_{14}H_{11}N_6O_2^+$ ,  $C_{12}H_{15}N_2O_5^+$ ,  $C_{12}H_{12}NO_5^+$ , and  $C_{11}H_{14}NO_3^+$  were observed both in the control and Biofield Energy Treated folic acid (Figure 2 & 3). The experimental data were well correlated with the published literature data [37].



**Figure 3:** Proposed fragmentation pattern of folic acid.

### Isotopic abundance ratio analysis

Both the control and Biofield Energy Treated folic acid samples showed the mass of a molecular ion at  $m/z$  441.92 (calcd for  $C_{19}H_{20}N_7O_6^+$ , 442.15) with 100% relative abundance in the spectra. The theoretical calculation of isotopic peak  $P_{M+1}$  for the protonated folic acid presented as below:

$$P(^{13}C) = [(19 \times 1.1\%) \times 100\% \text{ (the actual size of the } M^+ \text{ peak)}] / 100\% = 20.9\%$$

$$P(^2H) = [(20 \times 0.015\%) \times 100\%] / 100\% = 0.3\%$$

$$P(^{15}N) = [(7 \times 0.4\%) \times 100\%] / 100\% = 2.8\%$$

$$P(^{17}O) = [(6 \times 0.04\%) \times 100\%] / 100\% = 0.24\%$$

$$P_{M+1} \text{ i.e. } ^{13}C, ^2H, ^{15}N, \text{ and } ^{17}O \text{ contributions from } C_{19}H_{20}N_7O_6^+ \text{ to } m/z \text{ 442.92} = 24.24\%$$

The calculated isotopic abundance of  $P_{M+1}$  value 24.24% was higher to the experimental value (14.34%) (Table 1). From the above calculation, it has been found that  $^{13}C$  and  $^{15}N$  have the major contribution to  $m/z$  442.92.

The LC-MS based isotopic abundance ratio analysis  $P_M$  and  $P_{M+1}$  for folic acid near  $m/z$  441.92 and 442.92, respectively of the control and Biofield Energy Treated samples, which were obtained from the observed relative peak intensities of  $[M^+]$  and  $[(M+1)^+]$  peaks, respectively in the ESI-MS spectra (Table 1). The isotopic abundance ratio of  $P_{M+1}/P_M$  ( $^2H/^1H$  or  $^{13}C/^12C$  or  $^{15}N/^14N$  or  $^{17}O/^16O$ ) in Consciousness Energy Healing Treated folic acid was significantly increased by 69.8% compared to the control sample (Table 1). Thus, the  $^{13}C$ ,  $^2H$ ,  $^{15}N$ , and  $^{17}O$  contributions from  $C_{19}H_{20}N_7O_6^+$  to  $m/z$  442.75 in the Biofield Energy Treated sample was increased compared to the control sample.

**Table 1:** LC-MS based isotopic abundance analysis results in Biofield Energy Treated folic acid compared to the control sample.

Parameter	Control Sample	Biofield Energy Treated sample
$P_M$ at $m/z$ 441.92 (%)	100	100
$P_{M+1}$ at $m/z$ 442.92 (%)	14.34	24.35
$P_{M+1}/P_M$	0.14	0.24
% Change of isotopic abundance ratio ( $P_{M+1}/P_M$ ) with respect to the control sample		69.8

$P_M$ : the relative peak intensity of the parent molecular ion [ $M^+$ ];  $P_{M+1}$ : the relative peak intensity of the isotopic molecular ion [ $(M+1)^+$ ],  $M$ : mass of the parent molecule.

Our LC-MS study confirmed the structure of the sample as folic acid. The isotopic abundance ratio of  $P_{M+1}/P_M$  ( $^2H/^1H$  or  $^{13}C/^12C$  or  $^{17}O/^16O$ ) in the Biofield Energy Treated folic acid was significantly increased compared to the control sample. According to modern physics, neutrinos have the properties to change identities which are only possible if the neutrinos possess mass and have the ability to interchange their phase from one phase to another internally. Therefore, the neutrinos have the ability to interact with protons and neutrons in the nucleus, which indicated a close relationship between neutrino and the isotope formation [15,32,33]. The altered isotopic composition in the molecular level of The Trivedi Effect<sup>®</sup>-Consciousness Energy Healing Treated folic acid might have altered the neutron to proton ratio in the nucleus. It can be hypothesized that the changes in isotopic abundance could be due to changes in nuclei possibly through the interference of neutrino particles *via* The Trivedi Effect<sup>®</sup> - Consciousness Energy Healing Treatment. The Biofield Energy Treated folic acid would be more suitable for the prevention and treatment of various diseases such as embryonic defects, clinical depression, altered memory and brain function, leuco- and thrombocytopenia, megaloblastic anemia, cardiovascular disease, malignancies, neural tube defects, depression, allergic diseases, and lower bone density, etc.

## Conclusion

The Trivedi Effect<sup>®</sup>-Consciousness Energy Healing Treatment (Biofield Energy Treatment) showed the significant impact on the isotopic abundance ratio of folic acid (vitamin B<sub>9</sub> or folate). The LC-MS spectra of both control and Biofield Energy Treated samples at retention time ( $R_t$ ) 1.83 minutes exhibited the mass of the molecular ion peak at 441.92 (calcd for  $C_{19}H_{20}N_7O_6+$ , 442.15) along with low molecular fragmented mass peaks at  $m/z$  295.17, 267.17, 250.25, and 208 for  $C_{14}H_{11}N_6O_2+$ ,  $C_{12}H_{15}N_2O_5+$ ,  $C_{12}H_{12}NO_5+$ , and  $C_{11}H_{14}NO_3+$  were also observed. The isotopic abundance ratio of  $P_{M+1}/P_M$  ( $^2H/^1H$  or  $^{13}C/^12C$  or  $^{15}N/^14N$  or  $^{17}O/^16O$ ) in the Biofield Energy Treated folic acid was significantly increased by 69.80% compared with the control sample. Therefore, the  $^{13}C$ ,  $^2H$ ,  $^{15}N$ , and  $^{17}O$  contributions from  $C_{19}H_{20}N_7O_6+$  to  $m/z$  442.75 in the Biofield Energy Treated sample were significantly increased compared with the control sample. The isotopic abundance ratio of  $P_{M+1}/P_M$  ( $^2H/^1H$  or  $^{13}C/^12C$  or  $^{15}N/^14N$  or  $^{17}O/^16O$ ) in the Biofield Energy Treated folic acid was significantly increased compared to the control sample. It can be assumed that

the changes in isotopic abundance and mass peak intensities due to changes in nuclei possibly through the interference of neutrino particles *via* The Trivedi Effect<sup>®</sup> - Consciousness Energy Healing Treatment. The new form of Biofield Energy Treated folic acid would aid in designing novel pharmaceutical formulations that might offer better therapeutic response against glossitis, diarrhea, confusion, depression, anemia, gray hair, mouth sores, poor growth, swollen tongue, congenital deformities, embryonic defects, altered memory and brain function, leuco- and thrombocytopenia, cardiovascular disease, neural tube defects, and malignancies, risk of potentially developing allergic diseases, long-term risk of lower bone density, etc.

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