

Isotopic Abundance Ratio Analysis of Consciousness Energy Healing Treated Folic Acid

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ABSTRACT

Folic acid is the water-soluble vitamin used as a dietary supplement, involved in the synthesis and repair of DNA and RNA, aiding rapid cell division and growth, enhancing the overall brain functions, etc. in the body. In this research work, the impact of the Trivedi Effect[®] on the structural properties and the isotopic abundance ratio of folic acid was investigated using LC-MS spectroscopy. The folic acid power was divided into control and treated parts. Only the treated folic acid received the Trivedi Effect[®]-Consciousness Energy Healing Treatment remotely by a well-known Biofield Energy Healer, Mahendra Kumar Trivedi. The chromatograms of both the folic acid samples showed a major peak at retention time 1.8 min, and molecular ion mass peak at 441.92 along with low molecular fragmented mass peaks. The % peak area of the treated folic acid (76.25%) was significantly increased by 9.95% compared to the control sample (69.35%). The isotopic abundance ratio of P_{M+1}/P_M ($^2\text{H}/^1\text{H}$ or $^{13}\text{C}/^{12}\text{C}$ or $^{15}\text{N}/^{14}\text{N}$ or $^{17}\text{O}/^{16}\text{O}$) in the treated folic acid was significantly decreased by 62.76% compared with the control sample. Hence, the ^{13}C , ^2H , ^{15}N , and ^{17}O contributions from $\text{C}_{19}\text{H}_{20}\text{N}_7\text{O}_6^+$ to m/z 443 in the treated sample were significantly decreased compared with the control sample. The decrease in isotopic abundance might occur due to the changes in nuclei, possibly through the possible mediation of neutrino particles *via* the Trivedi Effect[®]-Consciousness Energy Healing Treatment. The decreased isotopic abundance ratio and peak area of the Consciousness Energy Healing Treated folic acid may decrease the chemical bond strength and decrease its stability but may increase its solubility and bioavailability in the body. The novel Biofield Energy Treated folic acid could be more efficacious against embryonic defects, megaloblastic anemia, clinical depression, cardiovascular disease, altered memory and brain function, leuco- and thrombocytopenia, malignancies, neural tube defects, depression, allergic diseases, and lower bone density, etc.

Keywords: The Trivedi Effect[®], Consciousness Energy Healing Treatment, Biofield Energy, Folic acid, LC-MS, Isotopic abundance

INTRODUCTION

Folic acid is a B vitamin (Vitamin B₉) used in food supplements [1]. Humans cannot prepare folate in the body itself, so it is required from the diets/supplements, making it an essential vitamin. The daily requirement of folate for adults is 400 micrograms, and for women in pregnancy is 400 to 800 mcg of folic acid a day [2]. The folic acid is mainly available in the food items of fruits, avocado, dark green leafy vegetables, nuts, soybeans, chickpeas, beetroot, asparagus, yeast, kale, liver, dairy products, meat, eggs, seafood, grains, and some beers [3-6]. Many roles played by folic acid in the body are very important, i.e., physicochemical reactions, enzymatic reactions included in vitamin metabolism, and amino acid synthesis. It also helps in the synthesis and repair of DNA and RNA, aiding rapid cell division and growth, enhancing the overall brain health, and in age-related hearing loss [7]. It also plays an important role in normal blood formation, immune function, amino

acid synthesis, maintain homocysteine levels, cell division, maintain maternal tissue growth during pregnancy, reduction of tiredness, and fatigue. Lack of folic acid in the body due to the inadequate intake of folate-rich food items, poor absorption due to Crohn's disease or celiac disease, abnormal metabolism, increased requirements during pregnancy or breastfeeding, genetic disorders, alcohol consumption, certain medicines like phenytoin, sulfasalazine, etc. reduce

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folic absorption, etc. Folic acid deficiency may be responsible for many diseases, i.e., glossitis, depression, confusion, anemia, grey hair, diarrhea, mouth sores, swollen tongue, poor growth, congenital deformities, embryonic defects, cardiovascular disease, altered memory and brain function, leuco- and thrombocytopenia, particular neural tube defects, and malignancies, risk of lower bone density, risk of developing allergic diseases, etc. [8-10].

The major issue associated with folic acid is very poor solubility in water, heat-sensitive, and decompose rapidly in the presence of light [11]. Thus, improvement of the physicochemical properties of folic acid is very important while preparing the nutraceutical and pharmaceutical formulations in the industries. The Trivedi Effect®-Consciousness Energy Healing Treatment scientifically proved that it has the capability to improve the physicochemical properties, isotopic abundance, and bioavailability of many pharmaceutical/nutraceutical compounds [12-14]. The Trivedi Effect® is a natural and only scientifically proven phenomenon in which an individual can harness this energy from the "Universe" and transfer it anywhere on the planet *via* the possible mediation of neutrinos [15]. "Biofield" is a unique and para-dimensional electromagnetic energy field that exists surrounding the living body. Energy Therapies are popular nowadays, which are accepted worldwide against various diseases and for better health and wellness [16-18]. Biofield Energy Healing therapy is recognized as a Complementary and Alternative Medicine (CAM) by the National Center of Complementary and Integrative Health (NCCIH) with other therapies, medicines, and practices such as Ayurveda, traditional Chinese herbs and medicines, homeopathy, hypnotherapy, Reiki, yoga, meditation, Tai Chi, Qi Gong, chiropractic/osteopathic manipulation, cranial sacral therapy, acupressure, acupuncture, healing touch, movement therapy, naturopathy, etc. [19]. CAM therapies have been adopted worldwide, whereas most of the U.S.A. people also adopted for better health and wellness [20]. The Trivedi Effect®-Consciousness Energy Healing Treatment also has a significant impact on the transformation of metals, ceramics, polymer, organic compounds, microorganisms, the productivity of crops, and cancer cells [21-30].

To understand the isotope effects resulting from the alterations of the isotopic composition, the study on the natural stable isotope ratio analysis has many applications in the different fields of sciences [31-33]. The sophisticated techniques like 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]. Therefore, the LC-MS based structural characterization and the isotopic abundance ratio in the Trivedi Effect®-Consciousness Energy Healing Treated folic acid were performed compared to the control sample.

MATERIALS AND METHODS

Chemicals and Reagents

The test sample folic acid power sample was purchased from Alfa Aesar, India. The solvents like methanol, acetonitrile, ammonium acetate was purchased from Merck, India.

Consciousness Energy Healing Treatment Strategies

The folic acid powder sample was divided into two parts and termed as control and treated parts. The control folic acid did not receive the Biofield Energy Treatment. But the sample was treated with a "sham" healer who did not have any knowledge about the Biofield Energy Treatment. However, the treated folic acid was received the Trivedi Effect®-Consciousness Energy Healing Treatment remotely for 3 min under standard laboratory conditions by a well-known Biofield Energy Healer, Mahendra Kumar Trivedi, USA. After the treatment, both the 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 both the folic acid samples was carried out with the help of LC-MS ThermoFisher Scientific, USA equipped with an ion trap detector connected with a triple-stage quadrupole mass spectrometer. Thermo Scientific Synchronis C18 (Length-250 mm X ID 4.6 mm X 5 micron) reversed phase column was used, which maintained at 25°C. The methanol and water (1:1) were used as a diluent for sample preparation. The injection volume was 10 µL and the analyte was eluted using acetonitrile +10 mM ammonium acetate (85:15) pumped at a constant flow rate of 1 mL/min (total run time was 10 min). Peaks were monitored at 220 nm using the PDA detector. The mass spectrometric analysis was performed under ESI +ve ion mode.

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 treated folic acid was calculated. The change in the isotopic abundance ratio was calculated using the following equation.

$$\begin{aligned} (\%) \text{Change in isotopic abundance ratio} &= [(IAR_{Treated} - IAR_{Control}) \\ & / IAR_{Control}] \times 100 \end{aligned}$$

Where $IAR_{Treated}$ and $IAR_{Control}$ were the isotopic abundance ratio in the treated and control folic acid.

RESULTS AND DISCUSSION

Liquid Chromatography-Mass Spectrometry (LC-MS)

The chromatograms of the folic acid samples showed the major peak at retention time (R_t) of 1.8 min (**Figure 1**). The % peak area near R_t 1.8 min of the Biofield Energy Treated folic acid (76.25%) was significantly increased by 9.95% compared to the control sample (69.35%). The increased peak area of the Biofield Energy Treated folic acid indicated that the solubility of folic acid was significantly increased after the Trivedi Effect[®]-Consciousness Energy Healing Treatment. The data was supported by one of the studies, in which the particle sizes are significantly decreased and the surface area was significantly increased [12]. The solubility

of folic acid is a major problem. Thus, the Biofield Energy Treated folic acid would be more soluble, bioavailable, and more efficacious nutraceutical and pharmaceutical formulations.

Both the samples exhibited the molecular ion of folic acid ($C_{19}H_{20}N_7O_6^+$) adduct with hydrogen ion at m/z 441.92 (calcd for $C_{19}H_{20}N_7O_6^+$, 442.15) along with the fragmented mass peaks at m/z 295.08, 267, 250.17, and 207.92 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 in mass spectra (**Figures 2 and 3**). The experimental data were well matched with the reported literature [37].

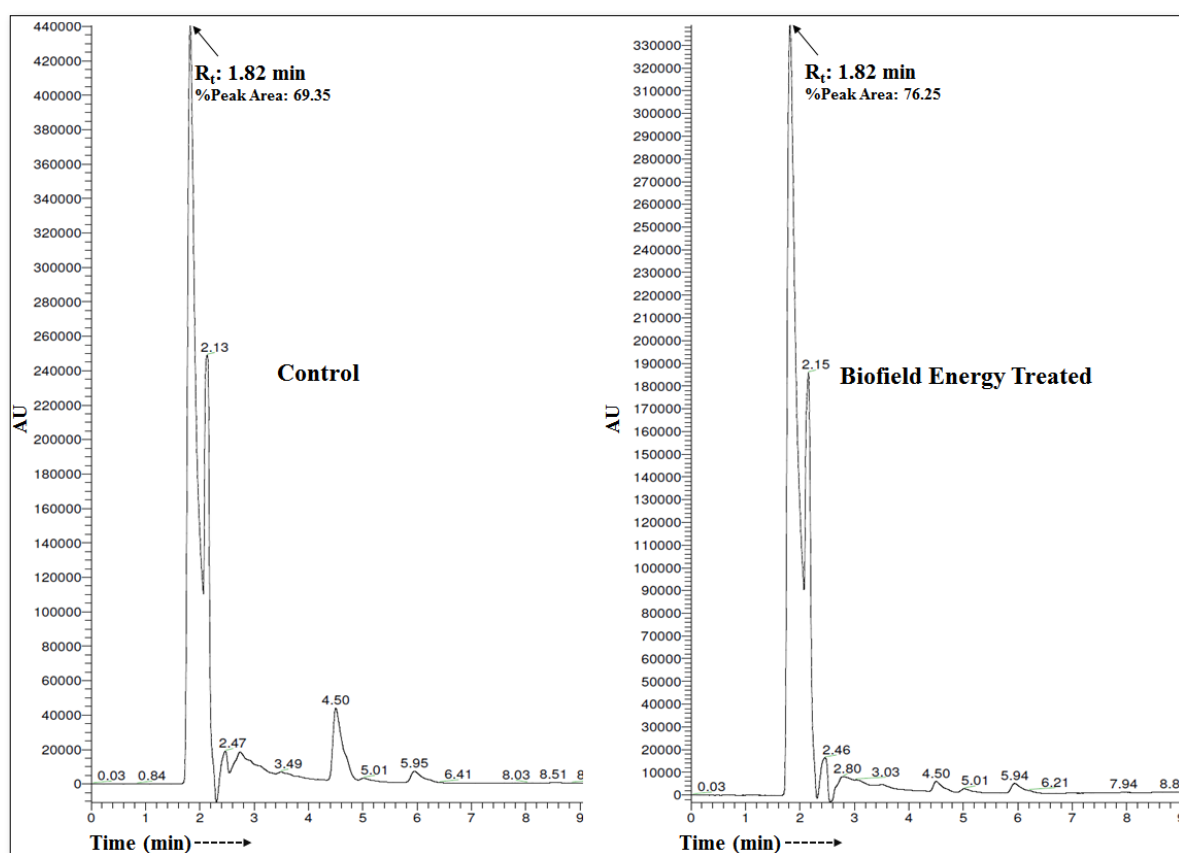


Figure 1. Liquid chromatograms of the control and Biofield Energy Treated folic acid.

Isotopic Abundance Ratio Analysis

The mass of a molecular ion at m/z 442 showed the 100% relative abundance in the mass 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} i.e., ^{13}C , 2H , ^{15}N , and ^{17}O contributions from $C_{19}H_{20}N_7O_6^+$ to m/z 442.92 = 24.24%

The calculated isotopic abundance of P_{M+1} value 24.24% was lower to the experimental value (43.13%) (**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 isotopic abundance ratio analysis P_M and P_{M+1} for folic acid at m/z 442 and 443, respectively of both the samples,

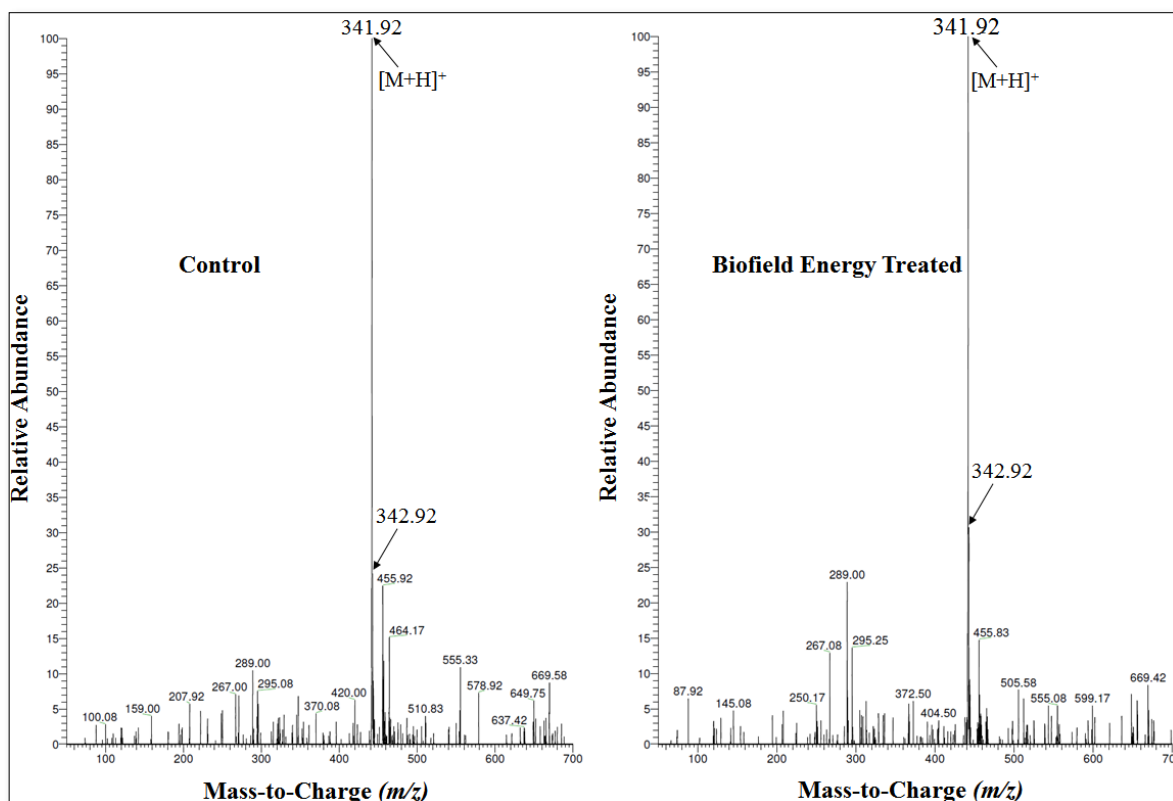


Figure 2. Mass spectra of the control and Biofield Energy Treated folic acid at R_t 1.8 min.

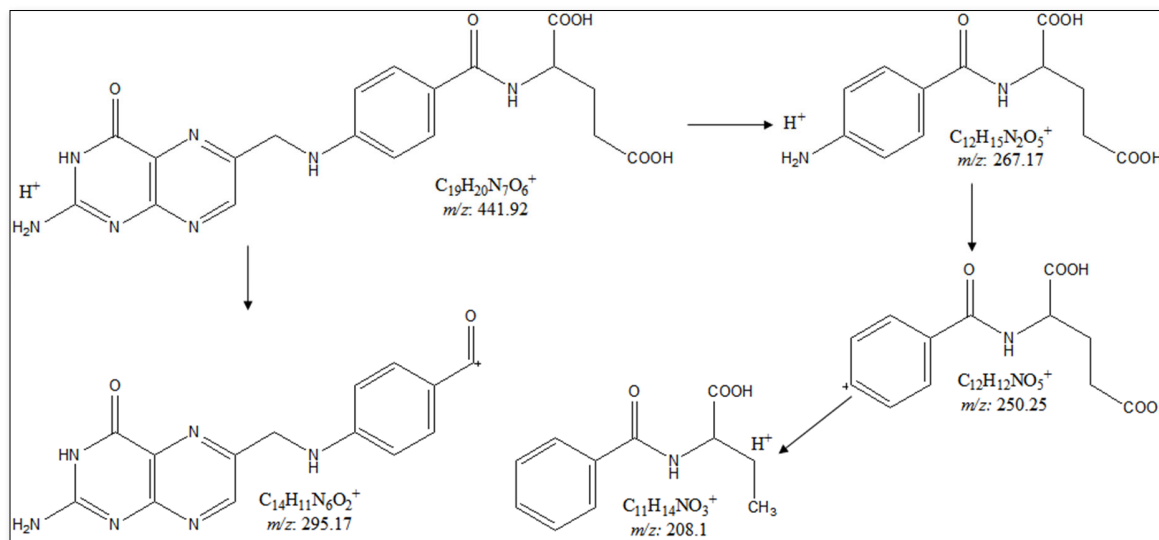


Figure 3. Proposed fragmentation pattern of folic acid.

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 $^{13}C/^{12}C$ or $^{15}N/^{14}N$ or $^{17}O/^{16}O$) in treated folic acid was significantly decreased by 62.76% 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 442.92 in the

treated folic acid was decreased compared to the control sample.

The isotopic abundance ratio of P_{M+1}/P_M ($^2H/^1H$ or $^{13}C/^{12}C$ or $^{17}O/^{16}O$) in the Biofield Energy Treated folic acid was significantly increased compared to the control sample. The altered isotopic composition in the Biofield Energy

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 442 (%)	100	100
P_{M+1} at m/z 443 (%)	43.13	16.06
P_{M+1}/P_M	0.43	0.16
% Change of isotopic abundance ratio (P_{M+1}/P_M) with respect to the control sample		-62.76

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

CONCLUSIONS

The Trivedi Effect[®]-Consciousness Energy Healing Treatment showed a significant impact on the isotopic abundance ratio of folic acid. The chromatograms of both the folic acid samples showed a major peak at retention time 1.8 min, and molecular ion mass peak at 441.92 along with low molecular fragmented mass peaks were also observed. The % peak area near R_t 1.8 min of the Biofield Energy Treated folic acid (76.25%) was significantly increased by 9.95% compared to the control sample (69.35%). 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 decreased by 62.76% compared with the control sample. Hence, the ^{13}C , 2H , ^{15}N , and ^{17}O contributions from $C_{19}H_{20}N_7O_6^+$ to m/z 443 in the Biofield Energy Treated sample were significantly decreased compared with the control sample. The decrease in isotopic abundance might occur due to the changes in nuclei possibly through the possible mediation of neutrino particles *via* the Trivedi Effect[®]-Consciousness Energy Healing Treatment. The decreased isotopic abundance ratio and peak area of the Consciousness Energy Healing Treated folic acid may decrease the chemical bond strength and decrease its stability, but may increase its solubility and bioavailability in the body. The novel Trivedi Effect[®]-Consciousness Energy Healing Treated folic acid could be more efficacious against embryonic defects, megaloblastic anemia, clinical depression, cardiovascular disease, altered memory and brain function, leuco- and thrombocytopenia, malignancies, neural tube defects, depression, allergic diseases, and lower bone density, etc.

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