



LC-MS Based Isotopic Abundance Ratio Analysis of the Consciousness Energy Treated L-Cysteine

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Abstract

L-cysteine is a non-essential amino acid but may be essential for new-borns, the elderly, and individuals with specific metabolic disease or malabsorption syndromes. In this study the impact of the Trivedi Effect[®] on the isotopic abundance ratios of L-cysteine using LC-MS analytical techniques were analysed. The test sample L-cysteine was divided into Control and the Biofield Energy Treated sample. The treated L-cysteine sample received Biofield Energy Treatment (the Trivedi Effect[®]) remotely for ~3 minutes by Mr. Mahendra Kumar Trivedi, who was located in the USA, while the test samples were located in the research laboratory in India. The LC-MS spectra of both the control and treated samples at retention time (R_t) 1.96 minutes exhibited the mass of the molecular ion peak adduct with hydrogen ion at 122 along with low molecular fragmented mass peaks were also observed. The peak area of the Biofield Energy Treated sample (1904766.03) was significantly increased by 4.94% compared with the control sample (1815060.18). The isotopic abundance ratios of P_{M+1}/P_M (²H/¹H or ¹³C/¹²C or ¹⁵N/¹⁴N or ¹⁷O/¹⁶O or ³³S/³²S) and P_{M+2}/P_M (³⁴S/³²S) in the treated L-cysteine was significantly increased by 25.12% and 14.35%, respectively compared with the control sample. Hence, the ¹³C, ²H, ¹⁵N, ¹⁷O, ³³S, and ³⁴S contributions from C₃H₈N₂O₂S⁺ to m/z 123 and 124 in the treated L-cysteine were significantly increased compared to the control sample. The changes in peak area and isotopic abundance ratios might be the cause of changes in nuclei, possibly through the interference of neutrino particles via the Trivedi Effect[®]-Consciousness Energy Treatment. The increased isotopic abundance ratio of the treated L-cysteine may increase the intra-atomic bond strength, increase its stability, and shelf-life. The novel Biofield Energy Treated L-cysteine might have increased the stability, solubility, bioavailability, and shelf-life compared to the control sample. The new form of treated L-cysteine would be a better and more stable precursor in the food, cosmetics, pharmaceuticals, personal-care products, additives to cigarettes (act as an expectorant), preventative or antidote for some of the negative effects of alcohol, acetaminophen overdose, clinically used ranging from baldness to psoriasis, excellent for the treatment of asthmatics by enabling them to stop theophylline and other medications, enhances the effect of topically applied silver, tin and zinc salts for preventing dental cavities. In the near future, this Biofield Energy Treated L-cysteine may play a better role in the treatment of diabetes, psychosis, cancer, and seizures.

Keywords: Biofield Energy; Consciousness Energy Treatment; L-cysteine; The Trivedi Effect[®]; LC-MS

Introduction

Cysteine is a non-essential amino acid but essential for new-borns, the elderly, and individuals with specific metabolic disease or malabsorption syndromes. The cysteine plenty available in milk, garlic, egg, meat, red peppers, onions, oats, broccoli, brussels sprout, wheat germ, sprouted lentils, etc. Industrially it is also prepared from animal feathers, hair, and even from chemical synthesis [1-3]. Cysteine is a semi-essential sulfur-containing amino acid found in nails, skin, hair, etc. in the body. It contains a thiol group and available as a chiral molecule with dextrorotation

(D) and levorotation (L) forms [1]. The sulfhydryl group of cysteine has numerous biological functions, *i.e.*, it acts, as a precursor to the antioxidant glutathione and iron-sulfur clusters, metal cofactors in enzymes, detoxification, protein synthesis, metabolic functions, collagen production, translation of messenger RNA molecules to produce polypeptides, etc. [1-6]. It is also a precursor in the food, pharmaceuticals, cosmetics, personal-care industries, additives to cigarettes (as an expectorant), preventative or antidote for some of the harmful effects of alcohol (*i.e.*, liver damage and hangover),

production of more wool from sheep, acetaminophen overdose, clinically used ranging from baldness to psoriasis, used for the treatment of asthma, enhances the effect of topically applied silver, tin and zinc salts for preventing dental cavities [1-9]. According to the research works, in the near future, cysteine may play an important role in the treatment of psychosis, diabetes, cancer, and seizures [10]. The stability of L-cysteine is an issue in the neutral or slightly alkaline aqueous solutions, which is oxidized to cystine by air, and on decomposition, it emits very toxic fumes of sulphur oxides and nitrogen oxides [6]. The Trivedi Effect®-Consciousness Energy Treatment has the amazing abilities to transform the characteristic properties of both living and non-living object(s) [11-15]. The Trivedi Effect® is a natural and only scientifically proven phenomenon in which an expert can harness this inherently intelligent energy from the "Universal Energy Field" and transmit it anywhere on the planet *via* the possible mediation of neutrinos [16]. An energy field generated around the body due to the continuous movement of the charged particles in the body known as a "Biofield". The object(s) received the "Energy Therapy" respond to a useful way is known as the Biofield Energy Treatment. There are several Biofield based Energy Therapies that are used nowadays against various disease conditions [17-19]. Biofield Energy therapy has been recognized worldwide as a Complementary and Alternative Medicine health care approach by the National Center of Complementary and Integrative Health with other therapies, medicines and practices such as Ayurvedic medicine, yoga, meditation, homeopathy, traditional Chinese herbs and medicines, naturopathy, chiropractic/osteopathic manipulation, Tai Chi, Qi Gong, acupressure, aromatherapy, acupuncture, hypnotherapy, Reiki, healing touch, cranial-sacral therapy, etc. [20]. These CAM therapies have been adopted by most of the U.S.A. population [21]. Similarly, the Trivedi Effect®-Consciousness Energy Treatment also been reported with significant impact on the properties of polymers, ceramics, organic compounds, metals, cancer cell line, microbes, improved skin health, bone health, improved agricultural crop yield, productivity, and quality, and altered the isotopic abundance ratio, improved bioavailability of pharmaceutical/nutraceutical compounds [22-37]. The analysis of the natural stable isotope has the importance of many applications to understand the isotope effects resulting from the alterations of the isotopic composition [38-40]. Liquid chromatography-mass spectrometry (LC-MS) analytical technique is the widely used analytical techniques for the analysis of isotope ratio with sufficient precision [39]. The Trivedi Effect®-Consciousness Energy Treatment could be an economical approach to alter the isotopic abundance of L-cysteine with improved physicochemical properties for the food, cosmetic, pharmaceutical/ nutraceutical, and other industries. Thus, this study was designed and evaluated the LC-MS based structural characterization and the isotopic abundance ratios in the Consciousness Energy Treated L-cysteine compared to the control sample.

Materials and Methods

Chemicals and Reagents

The test sample L-cysteine (> 98%) was purchased from Alfa Aesar, India. Other chemicals like methanol, acetonitrile, and ammonium acetate were purchased from Merck, India.

Consciousness Energy Treatment Strategies

The test sample L-cysteine powder was divided into two parts. One part did not receive the Biofield Energy Treatment and therefore considered as the Control L-cysteine. The second part of the test compound received the Energy of Consciousness Treatment remotely for ~3 minutes through the Unique Energy Transmission process by Mr. Mahendra Kumar Trivedi, who was located in the USA, while the test samples were located in the research laboratory in India, and it was labelled as the Biofield Energy Treated L-cysteine. On the other hand, the Control sample was subjected to "sham" healer, who did not have any knowledge about the Biofield Energy Treatment, under the similar laboratory conditions. After that, the Biofield Energy Treated and un-treated L-cysteine samples were kept in sealed conditions and characterized using LC-MS analytical technique.

Characterization

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

The LC-MS analysis of the L-cysteine 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. The used here was a reversed phase column Thermo Scientific Synchronis C18 (250 mm X 4.6 mm X 5 micron), maintained at 25°C. The diluent used for the sample preparation was methanol. The L-cysteine solution injection volume was 20 µL and the analyte was eluted using acetonitrile (92%) + 0.1% ammonium acetate (8%) pumped at a constant flow rate of 0.8 mL/min. Chromatographic separation was achieved using gradient condition and the total run time was 10 min. Peaks were monitored at 210 nm 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 was appeared in LC along with the full scan were recorded. The natural abundance of each isotope (C, H, N, O, and S) 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 [40-43]. The LC-MS based isotopic abundance ratios (P_{M+1}/P_M and P_{M+2}/P_M) for the control and Biofield Energy Treated L-cysteine ($C_3H_8NO_2S^+$) were calculated.

$$\% \text{ Change in isotopic abundance ratio} = \left[\frac{(IAR_{\text{Treated}} - IAR_{\text{Control}})}{IAR_{\text{Control}}} \right] \times 100$$

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

Result and Discussion

Liquid Chromatography-Mass Spectrometry (LC-MS)

L-cysteine showed a single peak at retention time (R_t) of 1.96 minutes in both the chromatograms (Figure 1). The peak area of the Biofield Energy Treated sample (1904766.03) was significantly increased by 4.94% compared with the control sample (1815060.18). It indicated that the solubility of the Biofield Energy Treated sample might have increased compared to the control sample. The finding was also supported by the published literature

data [12]. The mass spectra of both the samples at R_t of 1.96 minutes exhibited the presence of the molecular ion of L-cysteine (Figure 2) at m/z 122 (calcd for $C_3H_8NO_2S^+$, 122.03). Along with the molecular ion peak, low molecular fragmented mass peaks at m/z 105, 102, 87, 76, and 59 for $C_3H_5O_2S^+$, $C_3H_2O_2S^+$, $C_3H_5NO_2^{2+}$ or $C_3H_5NS^+$, $C_2H_6NO_2^+$, and $C_2H_3O_2^+$ were observed in case of both the samples (Figures 2, 3). The experimental data were well supported by the published literature [44]. The L-cysteine samples showed the mass of a molecular ion at m/z 122 (calcd for $C_3H_8NO_2S^+$, 122.03) with 100% relative abundance in the spectra. The theoretical calculation of isotopic peak P_{M+1} for the protonated L-cysteine presented as below:

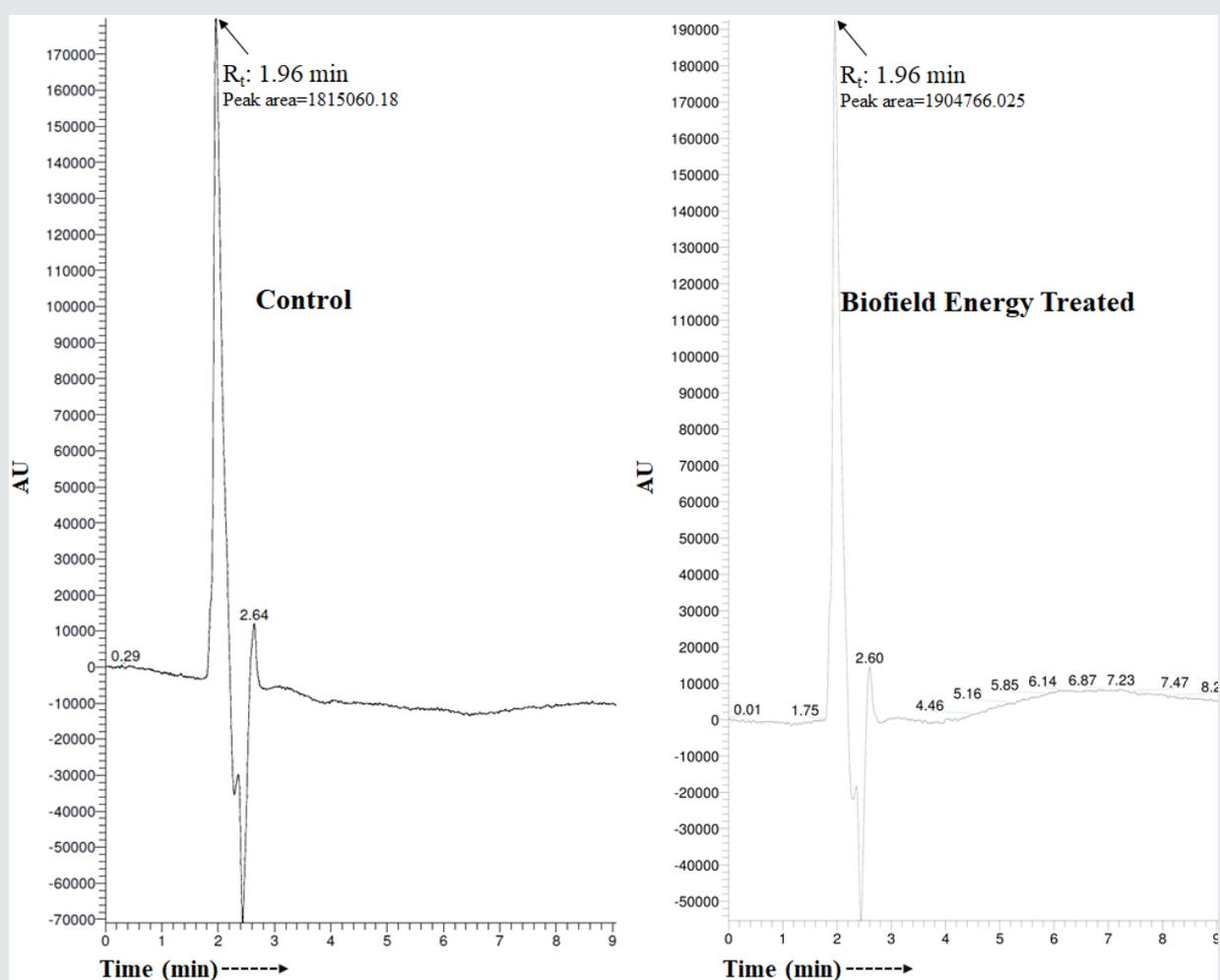


Figure 1: Liquid chromatograms of the control and Biofield Energy Treated L-cysteine.

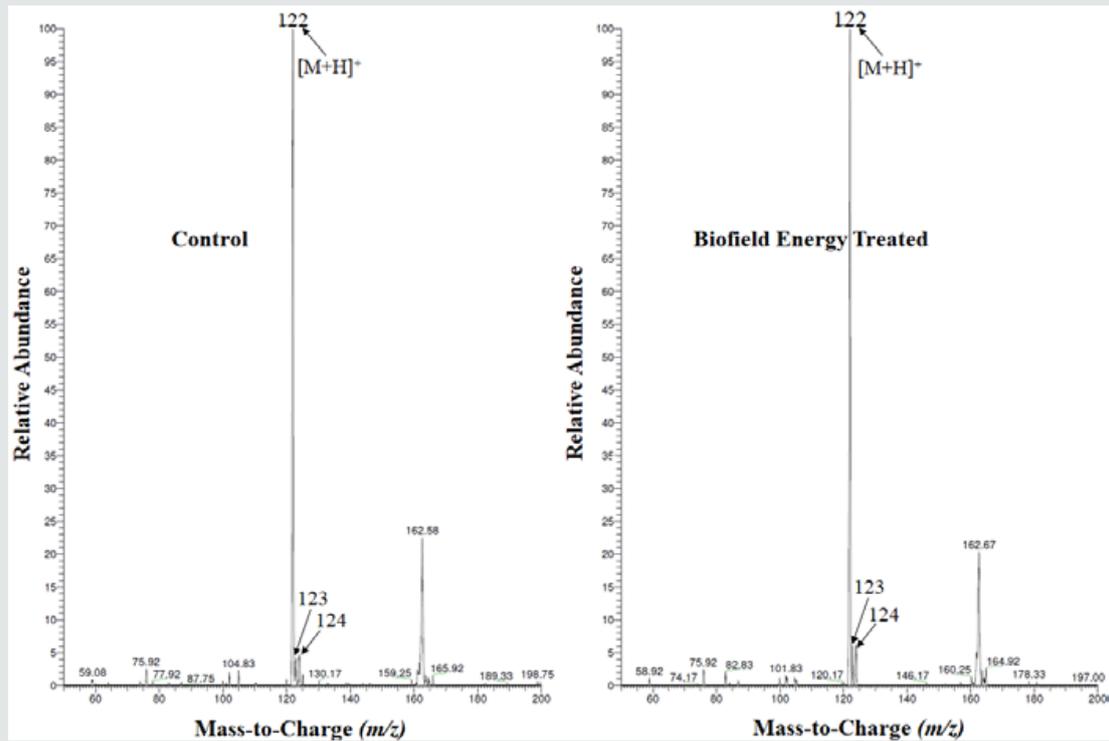


Figure 2: Mass spectra of the control and Biofield Energy Treated L-cysteine at R_t 1.96 minutes.

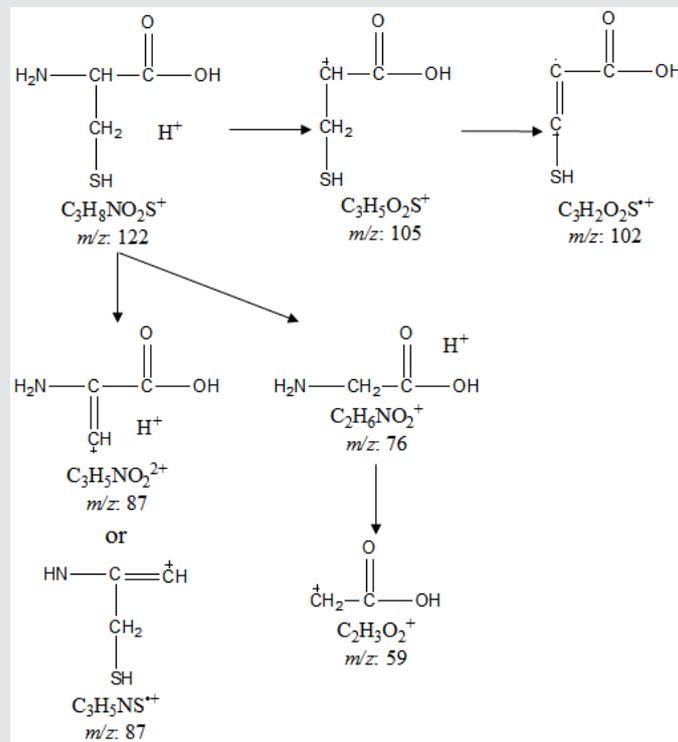


Figure 3: Proposed fragmentation pattern of L-cysteine.

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

Parameter	Control sample	Biofield Energy Treated sample
PM at m/z 122 (%)	100	100
PM+1 at m/z 123 (%)	4.3	5.38
PM+1/PM	0.043	0.0538
% Change of isotopic abundance ratio (P_{M+1}/P_M) with respect to the control sample		25.12
PM+2 at m/z 124 (%)	4.6	5.26
PM+2/PM	0.046	0.0526
% Change of isotopic abundance ratio (P_{M+2}/P_M) with respect to the control sample		14.35
PM: the relative peak intensity of the parent molecular ion [M^+]; P_{M+1} : the relative peak intensity of the isotopic molecular ion [$(M+1)^+$], P_{M+2} : the relative peak intensity of the isotopic molecular ion [$(M+2)^+$], M : mass of the parent molecule.		

$P(^{13}\text{C}) = [(3 \times 1.1\%) \times 100\% \text{ (the actual size of the } M^+ \text{ peak)}] / 100\% = 3.3\%$

$P(^2\text{H}) = [(8 \times 0.015\%) \times 100\%] / 100\% = 0.12\%$

$P(^{15}\text{N}) = [(1 \times 0.4\%) \times 100\%] / 100\% = 0.4\%$

$P(^{17}\text{O}) = [(2 \times 0.04\%) \times 100\%] / 100\% = 0.08\%$

$P(^{33}\text{S}) = [(1 \times 0.08\%) \times 100\%] / 100\% = 0.08\%$

P_{M+1} i.e. ^{13}C , ^2H , ^{15}N , ^{17}O , and ^{33}S contributions from $\text{C}_3\text{H}_8\text{NO}_2\text{S}^+$ to m/z 123 = 3.98%

Similarly, the theoretical calculation of P_{M+2} for L-cysteine was presented as below:

$P(^{34}\text{S}) = [(1 \times 4.21\%) \times 100\%] / 100\% = 4.21\%$

P_{M+2} i.e. ^{34}S contributions from $\text{C}_3\text{H}_8\text{NO}_2\text{S}^+$ to m/z 124 = 4.21%

The calculated isotopic abundance of P_{M+1} (3.98%) and P_{M+2} (4.21%) values was very close to the experimental values 4.3% and 4.6% (Table 1). From the above calculation, it has been found that ^{13}C , ^{15}N , and ^{34}S have the major contribution to m/z 123 and 124.

The isotopic abundance ratio analysis P_M , P_{M+1} , and P_{M+2} for L-cysteine near m/z 122, 123, and 124, respectively of both the samples were obtained from the observed relative peak intensities 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 ($^2\text{H}/^1\text{H}$ or $^{13}\text{C}/^{12}\text{C}$ or $^{15}\text{N}/^{14}\text{N}$ or $^{17}\text{O}/^{16}\text{O}$ or $^{33}\text{S}/^{32}\text{S}$) and P_{M+2}/P_M ($^{34}\text{S}/^{32}\text{S}$) in Consciousness Energy Treated L-cysteine was significantly increased by 25.12% and 14.35% compared to the control sample (Table 1). Thus, the ^{13}C , ^2H , ^{15}N , ^{17}O , ^{33}S , and ^{34}S contributions from $\text{C}_3\text{H}_8\text{NO}_2\text{S}^+$ to m/z 123 and 124 in the Biofield Energy Treated sample was significantly increased compared to the control sample.

The isotopic abundance ratios 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 $^{17}\text{O}/^{16}\text{O}$ or $^{33}\text{S}/^{32}\text{S}$) and P_{M+2}/P_M ($^{34}\text{S}/^{32}\text{S}$) in the Biofield Energy Treated L-cysteine were significantly increased compared to the control sample. The changes in isotopic abundance could be due to the possible interference of neutrino particles *via* the Trivedi Effect[®]-Consciousness Energy Treatment [16]. The altered isotopic

composition in the molecular level of the treated L-cysteine might have altered the neutron to proton ratio in the nucleus. A neutrino is an elementary particle that interacts through the weak subatomic force and gravity. 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 [39,40]. The isotopic abundance ratios $^2\text{H}/^1\text{H}$ or $^{13}\text{C}/^{12}\text{C}$ or $^{15}\text{N}/^{14}\text{N}$ or $^{17}\text{O}/^{16}\text{O}$ or $^{33}\text{S}/^{32}\text{S}$ or $^{34}\text{S}/^{32}\text{S}$ would influence the atomic bond vibration of treated L-cysteine [45]. The increased isotopic abundance ratio of the treated L-cysteine may increase the intra-atomic bond strength, increase its stability, and shelf-life. The novel Biofield Energy Treated L-cysteine might have increased the stability, solubility, bioavailability, and shelf-life compared to the control sample. The novel Biofield Energy Treated L-cysteine would be more important to the food, cosmetic, pharmaceutical/ nutraceutical, and other industries compared to the control sample.

Conclusion

The Trivedi Effect[®]-Consciousness Energy Treatment showed a significant impact on the chromatographic peak area and isotopic abundance ratio of L-cysteine. The LC-MS chromatographic peak area of the Biofield Energy Treated sample was significantly increased by 4.94% compared with the control sample. The isotopic abundance ratios 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 $^{17}\text{O}/^{16}\text{O}$ or $^{33}\text{S}/^{32}\text{S}$) and P_{M+2}/P_M ($^{34}\text{S}/^{32}\text{S}$) in the Biofield Energy Treated L-cysteine was significantly increased by 25.12% and 14.35%, respectively compared with the control sample. Hence, the ^{13}C , ^2H , ^{15}N , ^{17}O , ^{33}S , and ^{34}S contributions from $\text{C}_3\text{H}_8\text{NO}_2\text{S}^+$ to m/z 123 and 124 in the Biofield Energy Treated L-cysteine were significantly increased compared to the control sample. The changes in peak area and isotopic abundance ratios might be the cause of changes in nuclei possibly through the interference of neutrino particles *via* the Trivedi Effect[®]-Consciousness Energy Treatment. The increased isotopic abundance ratio of the Biofield Energy Treated L-cysteine may increase the intra-atomic bond strength, increase its stability, and shelf-life. The novel Biofield Energy Treated L-cysteine might have increased the stability, solubility, bioavailability, and shelf-life compared to the control sample. The new form of Biofield Energy

Treated L-cysteine would be a better and more stable precursor in the food, cosmetics, pharmaceuticals, personal-care products, additives to cigarettes, preventative or antidote for some of the negative effects of alcohol, acetaminophen overdose, clinically used ranging from baldness to psoriasis, excellent for the treatment of asthmatics by enabling them to stop theophylline and other medications, enhances the effect of topically applied silver, tin and zinc salts for preventing dental cavities. In the near future, this Biofield Energy Treated L-cysteine may play a better role in the treatment of diabetes, psychosis, cancer, and seizures.

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