



# Solid State Characterization of *Withania somnifera* (Ashwagandha) Root Extract After Treatment with Consciousness Energy Healing

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**Abstract:** Ashwagandha root extract is useful as an herbal medicine and nutraceuticals for the prevention and treatment of various diseases. The aim of the current study was to evaluate the influence of Consciousness Energy Healing Treatment (The Trivedi Effect<sup>®</sup>) on the physico-chemical, thermal and behavioral properties of ashwagandha root extract using powder X-ray diffraction (PXRD), particle size distribution analysis (PSD), Fourier transform infrared (FT-IR) spectrometry, UV-Vis spectroscopy, thermogravimetric analysis (TGA), and differential scanning calorimetry (DSC). Ashwagandha root extract was divided into two parts – one part was control without any Biofield Energy Treatment, and another part was treated with the Consciousness Energy Healing Treatment remotely by twenty renowned Biofield Energy Healers and defined as Biofield Energy Treated sample. The PXRD analysis concluded that both the control and treated samples were amorphous in nature. The particle size values at  $d_{10}$ ,  $d_{50}$ , and  $d_{90}$  of the treated sample were significantly decreased by 36.78%, 15.18%, and 5.06%, respectively compared with the control sample. Likewise, the surface area of the treated sample was significantly increased by 85.14% compared to the control sample. FT-IR results showed a small impact of Consciousness Energy Healing Treatment on the phytoconstituents of ashwagandha root extract to reduce the force constant of O-H (str.) bond. UV-vis analysis revealed that the wavelength for the maximum absorbance ( $\lambda_{max}$ ) of both the samples was at 206.4 in methanol. TGA revealed the three steps of thermal degradation and the total weight loss was decreased by 0.73% in the treated sample compared to the control sample. Consequently, the maximum thermal degradation temperature was found at 272.53°C and 393.35°C for two broad peaks in the treated sample was increased by 0.05% and 0.08%, respectively compared to the control the sample (272.67°C and 393.66°C). The DSC analysis indicated that the evaporation temperature and latent heat of vaporization were lowered significantly by 4.98% and 35.67%, respectively in the treated sample compared with the control sample. The current outcomes suggested that the Energy of Consciousness Healing Treatment might have the amazing capacity to enhance the solubility, dissolution, absorption, bioavailability and thermal stability of ashwagandha root extract in the various form of pharmaceutical and nutraceutical formulation by modifying its particle size and surface area. Thus, the Biofield Energy Treated

ashwagandha root extract might provide better therapeutic response against inflammatory diseases, immunological disorders, sexual disorders, arthritis, stress, cancer, ageing, diabetes, and other chronic infections.

**Keywords:** *Withania somnifera*, Biofield Energy Healing Treatment, Consciousness Energy Healing, The Trivedi Effect<sup>®</sup>, PXRD, Particle Size, TGA, DSC

## 1. Introduction

*Withania somnifera* (ashwagandha) is often touted as the Indian ginseng and traditionally known as winter cherry [1]. It is widely used herbal medicine and nutraceuticals for the treatment of various diseases including nervous and sexual disorders, infectious diseases, diabetes, cancer, ulcer, immunological disorders, stress, arthritis, etc. Since from the ancient time, it is used as a tonic to rejuvenate the body, arrest the ageing process, boost the defense against infectious disorders and also promote the longevity [2-11]. Ashwagandha root extract contains a wide array of phytochemicals and used as a dietary supplement for the better quality of life [3]. The major active phytoconstituents of ashwagandha root extract are withanolides. The oxidation at various sites of withanolide structure is responsible for the structural deviations in different classes of withanolides [11-14]. Well known withanolides such as withaferin A, withanolide D, withanolide E, etc. show various pharmacological activities including antioxidant, immunomodulating, anticancer, antibacterial, antiepileptic, neuroprotective, adaptogenic, spermatogenic, antidepressant, anti-anxiety, hepatoprotective, anti-inflammatory, antiarthritic, antimicrobial, antiulcer, hypolipidemic, aphrodisiac, hypoglycaemic, radiosensitizing, etc. [11, 14-19]. So, a new proprietary herbomineral formulation was prepared that consisted of ashwagandha root extract along with mineral salts such as magnesium gluconate, zinc chloride, and sodium selenate, which can be used for the prevention and treatment of various human disorders.

From the ancient-time, the living force preserved by every living organisms that contributes the 'life' is defined as prana by the Hindus, *ki* by the Japanese and *qi* or *chi* by the Chinese. This is believed to co-relate with the soul, spirit, and mind. This hypothetical vital force is considered now as Bioenergetics Field. The Biofield Energy Field is a dynamic electromagnetic field existing surround of the human body. The Biofield Energy is infinite and paradimensional. It can freely flow between the human and environment that leads to the continuous movement or matter of energy [20, 21]. Thus, the human has the ability to harness energy from the earth, the "universal energy field" and transmit it to any living or nonliving object(s) around the globe. The objects always receive the energy and respond in a useful way. This process is known as Biofield Energy Healing Treatment [22, 23]. Biofield (Putative Energy Fields) based Energy Therapies are used worldwide to promote health and healing [24]. National Center of Complementary and Integrative Health (NCCIH) has been recognized and accepted Biofield Energy Healing as a Complementary and Alternative Medicine (CAM) health

care approach in addition to other therapies, medicines and practices such as natural products, yoga, deep breathing, meditation, Tai Chi, Qi Gong, chiropractic/osteopathic manipulation, special diets, massage, homeopathy, progressive relaxation, acupressure, guided imagery, acupuncture, relaxation techniques, hypnotherapy, pilates, healing touch, rolfing structural integration, movement therapy, mindfulness, Ayurvedic medicine, traditional Chinese herbs and medicines, essential oils, naturopathy, aromatherapy, cranial sacral therapy, Reiki, and applied prayer (as is common in all religions, like Hinduism, Christianity, Buddhism, and Judaism) [25]. The Consciousness Energy Healing Treatment (The Trivedi Effect<sup>®</sup>) has been extensively studied with significant outcomes in many scientific fields such as biotechnology [26, 27], cancer research [28], genetics [29, 30], microbiology [31-33], altered structure of the atom with respect to the various polymers, metals, ceramics, and chemicals in materials science [34-36], altered physico-chemical properties of organic compounds [37-39], nutraceuticals [40, 41], pharmaceuticals [42, 43], and improved overall growth and yield of plants in agricultural science [44, 45]. Herbal extracts and their formulations despite their outstanding *in vitro* results exhibited poor *in vivo* activity, because of their low lipid solubility or improper molecular size, causing in deprived absorption and thus poor bioavailability. According to the current study on the bioavailability of major withanolides of *Withania somnifera*, Devkar *et al.* demonstrated that the nonpolar and low molecular weight withanolides are highly permeable, whereas the high glycosylated and polar withanolides displayed less permeability in their *in vitro* absorption model system [46]. As ashwagandha has the outstanding nutrition and medicinal values, researchers are still working on to find out an optimal dose range for reproducing the desired effects in human as well as to determine the safe, effective and non-toxic dosage form [47]. The physicochemical properties such as particle size, surface area, crystalline structure, crystallite size, and thermal properties of a drug play a vital role in bioavailability as well as the stability of the drug during formulation, processing, packaging, and storage [48, 49]. Biofield Energy Treatment (The Trivedi Effect<sup>®</sup>) has been reported to change the specific surface area, particle size, crystalline, chemical and thermal properties of an atom/ion through the possible mediation of neutrinos [50]. By considering all these aspect, the objective of the current study was to determine whether The Trivedi Effect<sup>®</sup> - Energy of Consciousness Healing Treatment (Biofield Energy Healing) can change the physical, structural, and thermal properties of *Withania somnifera* root extract in such a way that might assist in the

improvement of the solubility and absorption and also help in designing of any suitable nutraceuticals/pharmaceuticals formulation. The physical, spectroscopic, and thermal properties of both control and the Biofield Energy Treated ashwagandha root extracts were evaluated using various analytical techniques include Fourier transform infrared (FT-IR) spectrometry, ultraviolet-visible (UV-vis) spectroscopy, powder X-ray diffraction (PXRD), particle size distribution analysis (PSD), thermogravimetric analysis (TGA), and differential scanning calorimetry (DSC).

## 2. Materials and Methods

### 2.1. Chemicals and Reagents

*Withania somnifera* (Ashwagandha) root hydroalcoholic extract was purchased from Sanat Product Ltd., India. All other chemicals used in the experiment were of analytical grade available in India.

### 2.2. Consciousness Energy Healing Treatment Strategies

*Withania somnifera* root extract was one of the components of the new proprietary herbomineral formulation, developed by our research team, and it was used *per se* as the test compound for the current study. The test compound was divided into two parts, one part of the test compound was treated with The Trivedi Effect® - Consciousness Energy Healing Treatment (Biofield Energy Healing Treatment) by renowned Biofield Energy Healers and defined as Biofield Energy Treated sample, while the second part of the test compound did not receive any sort of such treatment and defined as untreated or control ashwagandha root extract sample. The Trivedi Effect® treatment was provided by the group of twenty renowned Biofield Energy Healers who participated in this study and performed the Biofield Energy Treatment (The Trivedi Effect®) remotely. Fifteen Biofield Energy Healers were remotely located in the U. S. A., two in Canada, one in the UK, one in Australia, and one in Germany, while the test compound was located in the research laboratory of GVK Biosciences Pvt. Ltd., Hyderabad, India. The Trivedi Effect® - Consciousness Energy Healing Treatment was provided for 5 minutes through Healer's Unique Energy Transmission process remotely to the test compound under the laboratory conditions. None of the Biofield Energy Healers in this study visited the laboratory in person, nor had any contact with the compounds. Similarly, the control sample was subjected to "sham" healers for 5 minutes, under the same laboratory conditions. The sham healer did not have any knowledge about Biofield Energy Treatment. After that, the treated and untreated samples were kept in similar sealed conditions and characterized thoroughly by PXRD, PSD, FT-IR, UV-visible spectroscopy, TGA, and DSC analysis.

### 2.3. Characterization

#### 2.3.1. Powder X-ray Diffraction (PXRD) Analysis

The PXRD analysis was accomplished on PANalytical

X'Pert Pro powder X-ray diffractometer system. The X-ray of wavelength 1.54056 Å was used. The data was collected in the form of a chart of the Bragg angle (2θ) vs. intensity, and a detailed table containing information on peak intensity counts, d value (Å), relative intensity (%), full width half maximum (FWHM) (θ°). From the XRD results, the crystallite size (G) was calculated using X'Pert data collector and X'Pert high score plus processing software. A total of ~500 mg of the control and treated samples individually were used for the analysis and prepared by back loading technique using the sample preparation kit. The sample was spread on the holder ring in sufficient quantity to fill the ring cavity. It was then pressed down using powder press block and scrap the powder that was in surplus using a glass slide to get a densely packed specimen. Consequently, the bottom plate was placed onto the holder ring and clamp in position. The sample holder was then removed from the sample preparation table by turning it upside down. A smooth surface of the sample was obtained to ensure optimum results.

#### 2.3.2. Particle Size Distribution (PSD) Analysis

The average particle size and particle size distribution were analyzed using Malvern Mastersizer 2000, UK with a detection range between 0.01 μm to 3000 μm. The sample unit was filled with the dispersant medium and operated the stirrer at 2500 rpm. Alignment of the optics was done and the background measurement was taken. After the background measurement, the sample was added into the sample unit with constant monitoring the obscuration and stopped the addition of sample when the obscuration reached in between 15% to 20%. When the obscuration was stable, the measurement was taken twice and the average was taken of two measurements. The average histogram of the two measurements was recorded. Along with histogram, the data was presented in table format which include particle size (μm). Also, the values at below 10% level (d<sub>10</sub>), 50% level (d<sub>50</sub>), and 90% level (d<sub>90</sub>) were calculated from the histogram and the calculations such as surface area (m<sup>2</sup>/g) were done by using software Mastersizer 2000.

Percent change in particle size (d) for at below 10% level (d<sub>10</sub>), 50% level (d<sub>50</sub>), and 90% level (d<sub>90</sub>) was calculated using following equation (1):

$$\% \text{ change in particle size} = \frac{d_{\text{Treated}} - d_{\text{Control}}}{d_{\text{Control}}} \times 100 \quad (1)$$

Where, d<sub>Control</sub> and d<sub>Treated</sub> are the particle size (μm) for at below 10% level (d<sub>10</sub>), 50% level (d<sub>50</sub>), and 90% level (d<sub>90</sub>) of the control and treated samples, respectively.

Percent change in surface area (S) was calculated using following equation (2):

$$\% \text{ change in surface area} = \frac{S_{\text{Treated}} - S_{\text{Control}}}{S_{\text{Control}}} \times 100 \quad (2)$$

Where, S<sub>Control</sub> and S<sub>Treated</sub> are the surface area of the control and treated samples, respectively.

### 2.3.3. Fourier Transform Infrared (FT-IR) Spectroscopy

FT-IR spectroscopy of ashwagandha root extract was performed on Spectrum two (Perkin Elmer, USA) Fourier transform infrared spectrometer with the frequency array of 400-4000  $\text{cm}^{-1}$  by using pressed KBr disk technique.

### 2.3.4. Ultra Violet-Visible Spectroscopy (UV-Vis) Analysis

The UV-Vis spectral analysis was carried out using Shimadzu UV-2450 with UV Probe, Japan. The spectrum was recorded using 1 cm quartz cell that has a slit width of 1.0 nm taking methanol as solvent. The wavelength range chosen for recording the spectra was 190-800 nm. The absorbance spectra (in the range of 0.2 to 0.9) and wavelength of maximum absorbance ( $\lambda_{\text{max}}$ ) were recorded.

### 2.3.5. Thermal Gravimetric Analysis (TGA)

TGA analysis was performed using Instrument TGA Q50 (TA Instruments, USA) at a heating rate of 10°C/min from room temperature *i. e.* 30°C to 900°C under nitrogen atmosphere. A total of ~14 mg of sample was used for the analysis and was taken on the platinum pan. In TGA, the weight loss for each step was recorded in grams as well as in percent loss with respect to the initial weight. Also, the onset, endset, and peak temperature for each step were recorded in TGA.

Percent change in weight loss (W) was calculated using following equation (3):

$$\% \text{ change in weight loss} = \frac{[W_{\text{Treated}} - W_{\text{Control}}]}{W_{\text{Control}}} \times 100 \quad (3)$$

Where,  $W_{\text{Control}}$  and  $W_{\text{Treated}}$  are the weight loss of the control and The Trivedi Effect<sup>®</sup> treated samples, respectively.

### 2.3.6. Differential Scanning Calorimetry (DSC)

Analysis was performed using the DSC Q20 (TA Instruments, USA) differential scanning calorimeter. A total of ~4 mg sample was weighed and sealed in aluminum pan and equilibrated at 25°C and heated up to 450°C at the heating rate of 10°C/min under nitrogen gas as purge atmosphere with flow rate of 50 mL/min. The value for onset, endset, peak temperature, peak height (mJ or mW), peak area, and change in heat (J/g) for each peak were recorded.

Percent change in melting point (T) was calculated using following equation (4):

$$\% \text{ change in melting point} = \frac{[T_{\text{Treated}} - T_{\text{Control}}]}{T_{\text{Control}}} \times 100 \quad (4)$$

Where,  $T_{\text{Control}}$  and  $T_{\text{Treated}}$  are the melting point of the control and treated samples, respectively.

Percent change in latent heat of fusion ( $\Delta H$ ) was calculated using following equation (5):

$$\% \text{ change in latent heat of fusion} = \frac{[\Delta H_{\text{Treated}} - \Delta H_{\text{Control}}]}{\Delta H_{\text{Control}}} \times 100 \quad (5)$$

Where,  $\Delta H_{\text{Control}}$  and  $\Delta H_{\text{Treated}}$  are the latent heat of fusion of the control and treated samples, respectively.

## 3. Results and Discussion

### 3.1. Powder X-ray Diffraction (PXRD) Analysis

PXRD study of the control and Consciousness Energy Healing Treated ashwagandha root extract was carried out to examine the crystalline pattern. Figure 1 showed the PXRD diffractograms of the control and Biofield Energy Treated ashwagandha root extract, which did not contribute any diffraction peaks. It was then concluded that both samples are amorphous in nature and the Consciousness Energy Healing Treatment has no effect compared to the control sample.

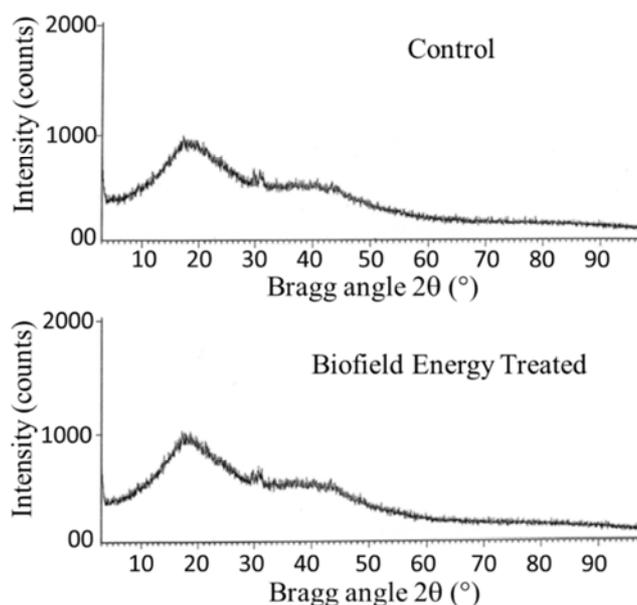


Figure 1. PXRD diffractograms of the control and Biofield Energy Treated *W. somnifera* (Ashwagandha) root extract.

### 3.2. Particle Size Distribution (PSD) Analysis

PSD analysis of the control and Consciousness Energy Healing Treated ashwagandha root extract was executed and the results are presented in Table 1. The particle size values of the Biofield Energy Treated ashwagandha root extract at  $d_{10}$ ,  $d_{50}$ , and  $d_{90}$  were found to be significantly decreased by 36.78%, 15.18%, and 5.06%, respectively compared with the control sample (Table 1). Correspondingly, the surface area of the Biofield Energy Treated ashwagandha root extract (0.077  $\text{m}^2/\text{g}$ ) was significantly increased by 85.14% compared with the control sample (0.137  $\text{m}^2/\text{g}$ ).

Table 1. Particle size ( $d_{10}$ ,  $d_{50}$ , and  $d_{90}$ ) and surface area of the control and Biofield Energy Treated *W. somnifera* (Ashwagandha) root extract.

Parameter	$d_{10}$ ( $\mu\text{m}$ )	$d_{50}$ ( $\mu\text{m}$ )	$d_{90}$ ( $\mu\text{m}$ )	Surface area ( $\text{m}^2/\text{g}$ )
Control	73.667	185.972	368.666	0.074
Biofield Energy Treated	46.571	157.745	350.015	0.137
Percent change (%) <sup>*</sup>	-36.78	-15.18	-5.06	85.14

\*denotes the percentage change in the particle size ( $d_{10}$ ,  $d_{50}$ , and  $d_{90}$ ) and surface area of the Biofield Energy Treated sample with respect to the control sample.

The results indicated that there might be an effect of Consciousness Energy Healing Treatment in the intrinsic physico-chemical properties of ashwagandha root extract. Many literature mentioned that particle size, shape and surface area of the nutraceuticals/pharmaceuticals have an impact on solubility, dissolution rate, *in vivo* bioavailability, dose uniformity and therapeutic efficacy as well as assist in the design of the new drug delivery systems [48, 51]. Decrease in size of the particle, and increase in the surface area increase the solubility of the solid particles as well as enhance the dissolution rate and bioavailability [52]. Thus, it is anticipated that The Trivedi Effect® - Consciousness Energy Healing Treated ashwagandha root extract might be absorbed in a faster rate and thus, can provide better bioavailability than the untreated sample.

### 3.3. Fourier Transform Infrared (FT-IR) Spectroscopy

The FT-IR spectra of the control and Biofield Energy Treated ashwagandha root extract are presented in Figure 2. The FT-IR vibrational frequency of the control and Biofield

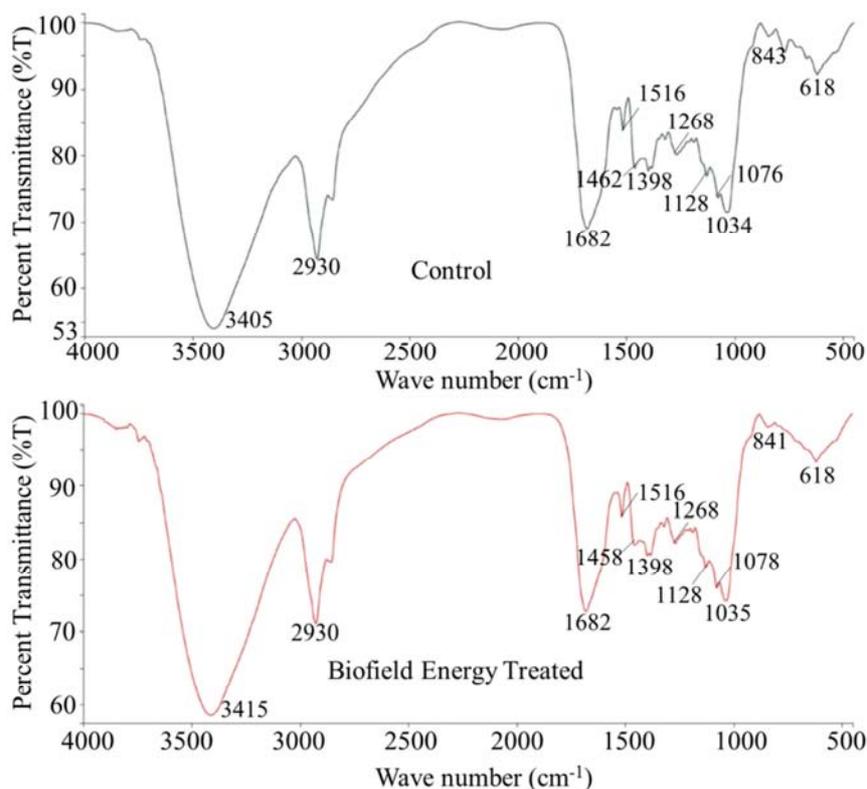


Figure 2. FT-IR spectra of the control and Biofield Energy Treated *W. somnifera* (Ashwagandha) root extract.

It has been observed from the FT-IR spectral data, that the vibrational frequencies for the C-H, C-C, C-O, and C=O groups of the Biofield Energy Treated ashwagandha were very close to the control sample (Figure 2; Table 2, entry 1-6). The O-H stretching of the control sample was at 3405 cm<sup>-1</sup>, and shifted upward to 3415 cm<sup>-1</sup> in the Biofield Energy Treated sample. It can be assumed that the force constant of O-H functional group in the Biofield Energy Treated sample was increased compared with the control sample. The broad

Energy Treated sample are presented in Table 2. The wavenumber of the absorbance ( $\nu$ ) of a diatomic can be calculated from the following equation (6) derived from Hooke's law:

$$\nu = \frac{1}{2\pi c} \sqrt{\frac{f(m_1 + m_2)}{m_1 m_2}} \quad (6)$$

Where,  $\nu$  = vibrational frequency (cm<sup>-1</sup>),  $c$  = the velocity of light (cm/s),  $m_1$  and  $m_2$  = the mass of atoms 1 and 2, respectively in g,  $f$  = the force constant of the bond (dyne/cm).

In equation (6) if other factors remain constant, the vibrational frequency (wavenumber) is directly proportional to the force constant *i. e.* for a certain functional group (for *e.g.* - C=O), changes in the vibrational frequency (wavenumber) indicate the alteration of the force constant. Several factors such as hybridization, bond strength, resonance, conjugation, *etc.* can affect the force constant [53, 54].

peak at 3600-3200 cm<sup>-1</sup> may be due to the O-H functional group frequency of the hydroxyl groups present in phytoconstituents present in the ashwagandha root extract. The presence of epoxide, unsaturated lactone, 1-keto-2-ene functions plays a vital role to elicit the pharmacological activities of withanolides [14, 55-57]. In summary, FT-IR results suggest that there was a little impact of Consciousness Energy Healing Treatment on *W. somnifera* at the atomic level to reduce the force constant of O-H (str.) bond.

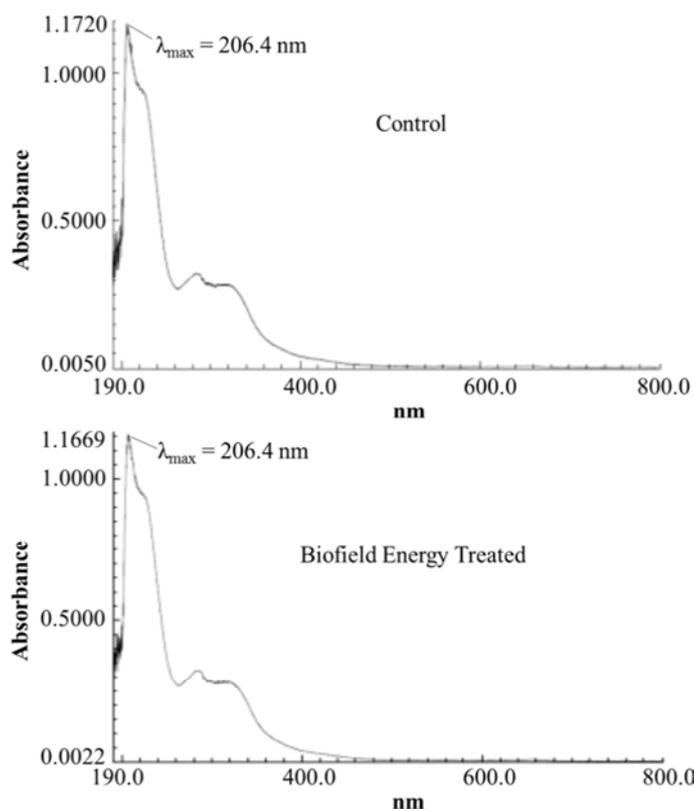
**Table 2.** FT-IR data of the control and Biofield Energy Treated *W. somnifera* (Ashwagandha) root extracts.

Entry No.	Mode of vibration	Characteristic absorption (s) of ashwagandha root extract (cm <sup>-1</sup> )	
		Control	Biofield Energy Treated
1	O-H stretching	3405	3415
2	C-H stretching	2930	2930
3	C=O stretching ( $\alpha$ , $\beta$ -unsaturated ketone)	1682	1682
4	C-C stretching	1516, 1462, 1398, 1268	1516, 1458, 1398, 1268
5	C-O in alkoxy	1128, 1076, 1034	1128, 1078, 1035
6	C-H aromatic bending	843, 618	841, 618

### 3.4. Ultraviolet-Visible Spectroscopy (UV-Vis) Analysis

The UV-vis spectra of the control and Biofield Energy Treated ashwagandha root extract are shown in Figure 3. The UV spectrum of the control and Biofield Energy Treated sample showed the maximum absorbance at 206.4 nm ( $\lambda_{\max}$ ). The peak at 206.4 nm was showed a minor shift of absorbance maxima from 1.1723 in control to 1.1671 in the Biofield Energy Treated sample. The UV absorbance occurs due to the different type of energy transitions from the singlet to the singlet excited state such as  $\sigma \rightarrow \sigma^*$ ,  $n \rightarrow \pi^*$ , and  $\pi \rightarrow \pi^*$ . These type of electronic

transitions are happen when the difference in energy between the lowest unoccupied molecular orbital and the highest occupied molecular orbital is significantly higher than the activation energy of the compound [59]. It has been reported that the wavelength for the maximum absorbance for the ashwagandha root extract was at 208.50 nm. Thus, no difference in the  $\lambda_{\max}$  of the Consciousness Energy Healing Treated sample was observed compared to the control sample. Hence, it is predicted that the structure of the phytoconstituents in the treated sample remained unaffected as compared with the control sample.

**Figure 3.** UV-vis spectra of the control and Biofield Energy Treated *W. somnifera* (Ashwagandha) root extracts.

### 3.5. Thermal Gravimetric Analysis (TGA)

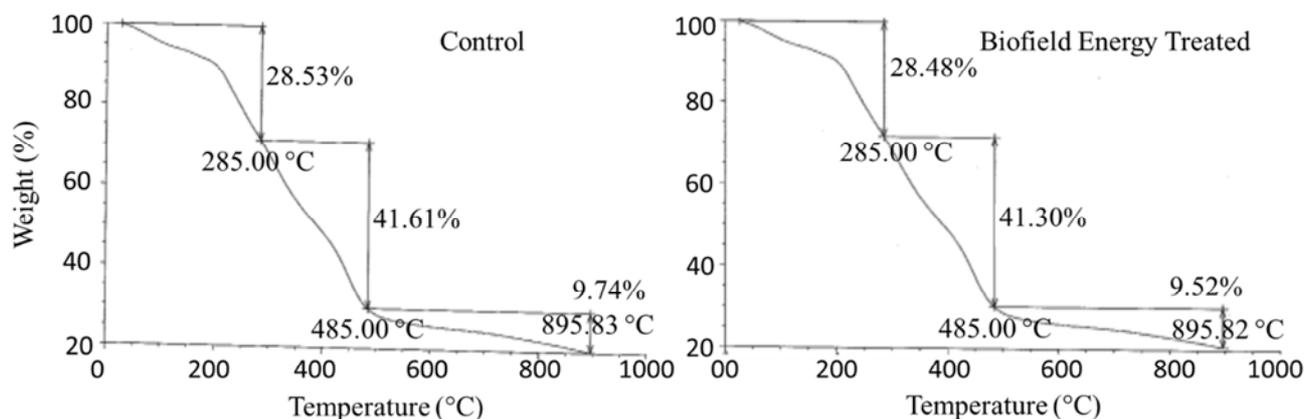
The thermal stability of the control and Biofield Energy Treated ashwagandha root extracts was carried out by means of TGA. The TGA thermograms showed three steps of thermal degradation as mentioned in Table 3 and Figure 4. In the 1<sup>st</sup>, 2<sup>nd</sup> and 3<sup>rd</sup> steps of thermal degradation, the weight loss of the

Biofield Energy Treated sample was decreased by 0.18%, 0.75%, and 2.26%, respectively compared with the control sample (Table 3). However, the total weight loss of the Biofield Energy Treated sample (79.30%) was reduced by 0.73% compared with the control sample (79.88%). It is then predicted that Consciousness Energy Healing Treatment might enhance the thermal stability of ashwagandha root extract.

**Table 3.** Thermal degradation steps of the control and Biofield Energy Treated *W. somnifera* (Ashwagandha) root extract.

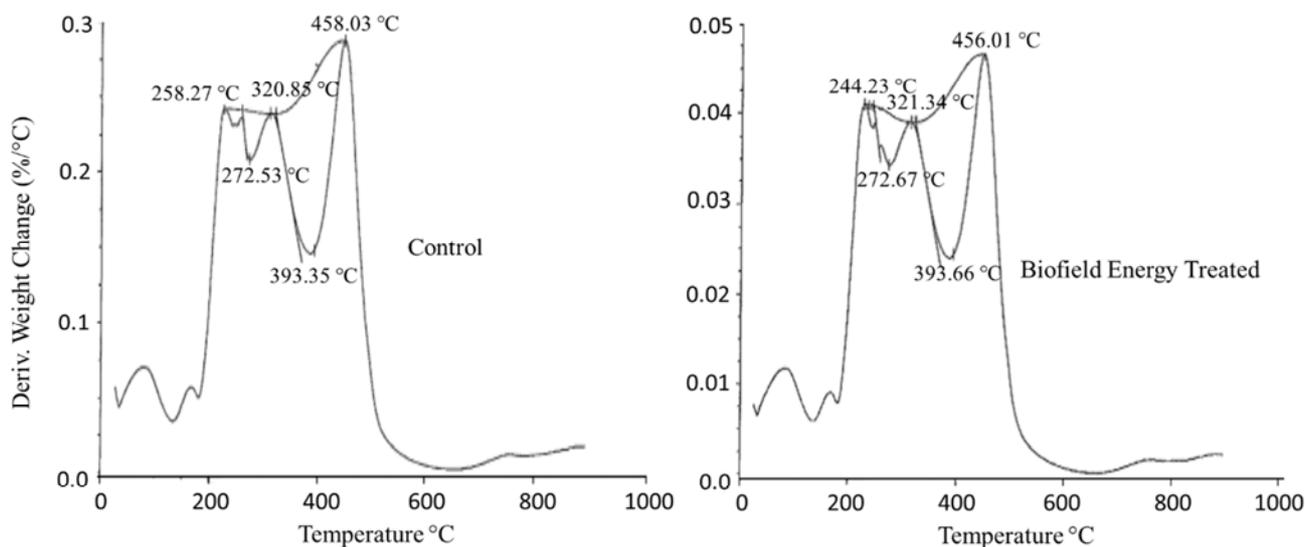
S. No.	Temperature (°C)		% Weight loss		% Change
	Control	Treated	Control	Treated	
1 <sup>st</sup> step of degradation	285.00	285.00	28.53	28.48	-0.18
2 <sup>nd</sup> step of degradation	485.00	485.00	41.61	41.30	-0.75
3 <sup>rd</sup> step of degradation	895.83	895.85	9.74	9.52	-2.26
Total weight loss			79.88	79.30	-0.73

\*denotes the percentage change in the weight loss of the Biofield Energy Treated sample with respect to the control sample.

**Figure 4.** TGA thermograms of the control and Biofield Energy Treated *W. somnifera* (Ashwagandha) root extract.

Similarly, the DTG thermograms of the control and Biofield Energy Treated ashwagandha root extract displayed two broad peaks (Figure 5). The maximum thermal degradation temperature at 272.53°C and 393.35°C for two broad peaks in the Biofield Energy Treated ashwagandha root extract was increased by 0.05% and 0.08%, respectively

compared to the control the sample (272.67°C and 393.66°C). Overall, the results indicated that the thermal stability of the Consciousness Energy Healing Treated ashwagandha was slightly increased compared to the control sample.

**Figure 5.** The DTG thermograms of the control and Biofield Energy Treated *W. somnifera* root extract.

### 3.6. Differential Scanning Calorimetry (DSC) Analysis

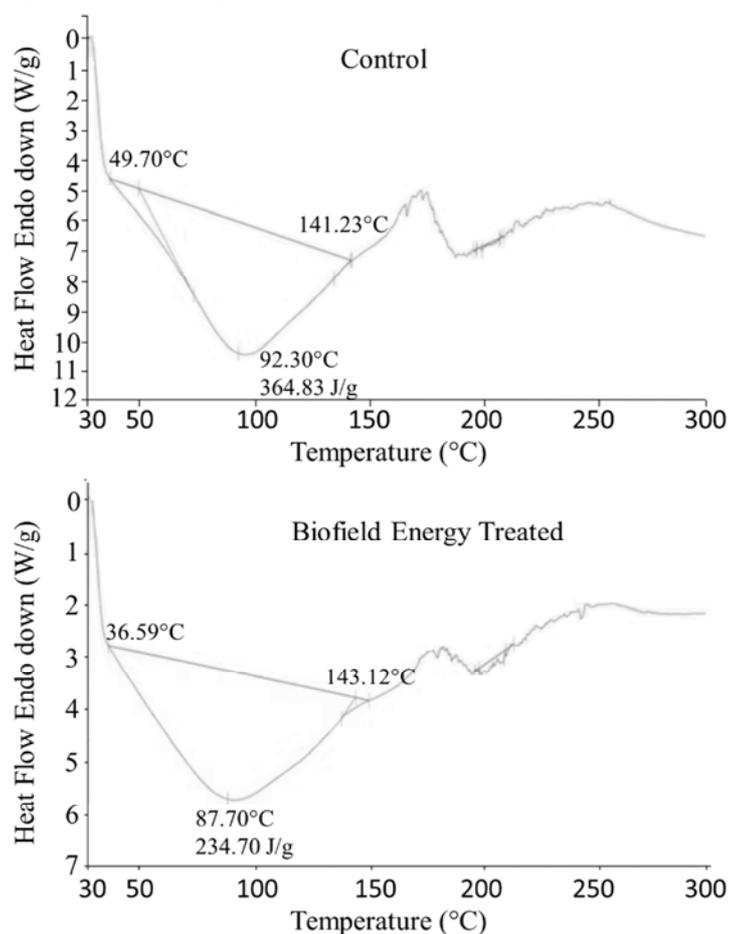
The DSC thermograms of the control and Consciousness Energy Healing Treated ashwagandha root extract are presented in Figure 6. The DSC thermograms of the control

and Consciousness Energy Healing Treated samples indicated the presence of a broad endothermic inflection at 92.30°C and 87.70°C, respectively. This might be the evaporation of the bounded water present in the ashwagandha root extract. This evaporation temperature and latent heat of

vaporization were decreased significantly by 4.98%, and 35.67%, respectively in the Consciousness Energy Healing Treated sample compared with the control sample (Table 4).

It is assumed that the Consciousness Energy Healing Treatment might influenced the intermolecular force in the Biofield Energy Treated sample, which probably decreases the heat change. The result suggests that the temperature for

water removal of the Biofield Energy Treated sample was decreased compared to the control sample. However, Biofield Energy Treated and control ashwagandha root extract indicated several endothermic peaks around 200°C (Figure 6), which might be due to the multiple phytoconstituents present in the root extract in a very small concentration [60, 61].



**Figure 6.** DSC thermograms of the control and Biofield Energy Treated *W. somnifera* (Ashwagandha) root extract.

**Table 4.** The melting point (°C) and latent heat of fusion (J/G) values of the control and Biofield Energy Treated *W. somnifera* (Ashwagandha) root extract.

Sample	( $T_{\text{onset}}$ ) °C	( $T_{\text{peak}}$ ) °C	( $T_{\text{endset}}$ ) °C	( $\Delta H_{\text{vaporization}}$ ) J/g
Control	49.70	92.30	141.23	364.83
Biofield Energy Treated	36.59	87.70	143.12	234.70
% Change*	-26.38	-4.98	1.34	-35.67

$T_{\text{onset}}$ : Onset vaporization temperature,  $T_{\text{peak}}$ : Peak vaporization temperature,  $T_{\text{endset}}$ : Endset vaporization temperature,  $\Delta H$ : Latent heat of vaporization, \*denotes the percentage change of the Biofield Energy Treated sample with respect to the control sample.

## 4. Conclusions

The current study revealed that Consciousness Energy Healing Treatment (The Trivedi Effect<sup>®</sup>) has the significant effect on the surface area, particle size, and thermal stability of the ashwagandha root extract. The PXRD analysis concluded that both the control and treated samples were amorphous in nature. The particle size values at  $d_{10}$ ,  $d_{50}$ , and  $d_{90}$  of the treated sample were significantly decreased by 36.78%, 15.18%, and 5.06%, respectively compared with the

control sample. Likewise, the surface area of the treated sample was significantly increased by 85.14% compared to the control sample. FT-IR results showed a small impact of Consciousness Energy Healing Treatment on the phytoconstituents of ashwagandha root extract at the atomic level to reduce the force constant of O-H (str.) bond. UV-vis analysis revealed that the wavelength for the maximum absorbance ( $\lambda_{\text{max}}$ ) of both the samples was at 206.4. TGA data revealed three steps of thermal degradation and the total weight loss was decreased by 0.73% in the treated sample compared to the control sample. Consequently, the maximum

thermal degradation temperature at 272.53°C and 393.35°C for two broad peaks in the treated sample was increased by 0.05% and 0.08%, respectively compared to the control the sample (272.67°C and 393.66°C). The DSC analysis indicated that the evaporation temperature and latent heat of vaporization were significantly decreased by 4.98% and 35.67%, respectively in the treated sample compared with the control sample. In summary, Energy of Consciousness Healing Treated ashwagandha root extract could have better solubility, absorption, dissolution, bioavailability, and long-term storage stability compared with the control sample and could be suitable for any pharmaceutical and nutraceutical formulation which might be providing better therapeutic response against various diseases such as allergies and septic shock, diabetes mellitus, stress-related disorders like anxiety, insomnia, depression, sleep disorder, mental restlessness (mind chattering), brain fog, impotency, lack of motivation, low libido, mood swings, confusion, migraines, headaches, forgetfulness, overwhelm, fear of the future, indecisiveness, worthlessness, frustration, irritability, loneliness, obsessive/compulsive behavior, chronic fatigue, and panic attacks; immunological and inflammatory diseases disorders like Hashimoto Thyroiditis, Diabetes, Lupus, Systemic Lupus Erythematosus, Chronic peptic ulcers, Asthma, Chronic active hepatitis, Tuberculosis, Hepatitis, Addison Disease, Graves' Disease, Pernicious, Crohn's disease, Rheumatoid arthritis, Irritable Bowel Syndrome, Aplastic Anemia, Multiple Sclerosis, Chronic periodontitis, Ulcerative colitis, Chronic sinusitis, Atherosclerosis, Myasthenia Gravis, Diverticulitis, Dermatitis, Arthritis, Psoriasis, Alopecia Areata, Scleroderma, Chronic Fatigue Syndrome, aging-related diseases like cardiovascular disease, arthritis, cancer, dementia, cataracts, Alzheimer's disease, osteoporosis, diabetes, glaucoma, hearing loss, hypertension, Parkinson's Disease, Motor Neurone Disease, Prion Disease, Spinocerebellar Ataxia, Friedreich's Ataxia, Spinal muscular atrophy, Lewy Body Disease, chronic infections, and many more.

## Abbreviations

DSC: Differential scanning calorimetry, FT-IR: Fourier transform infrared spectroscopy, HOMO: Highest energy occupied molecular orbital, LUMO: Lowest energy unoccupied molecular orbital, TGA: Thermal gravimetric analysis,  $T_{peak}$ : Peak vaporization temperature,  $\Delta H$ : Latent heat of vaporization, UV-vis: Ultraviolet-visible spectroscopy, PSD: Particle size distribution; PXRD: Powder X-ray diffraction.

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## References

- [1] Mirjalili MH, Moyano E, Bonfill M, Cusido RM, Palazón J (2009) Steroidal lactones from *Withania somnifera*, an ancient plant for novel medicine. *Molecules* 14: 2373-2393.
- [2] Singh N, Bhalla M, Jager P, Gilca M (2011) An overview on ashwagandha: A rasayana (rejuvenator) of ayurveda. *Afr J Tradit Complement Altern Med* 8: 208-213.
- [3] Alam N, Hossain M, Mottalib MA, Sulaiman SA, Gan SH, Khalil MI (2012) Methanolic extracts of *Withania somnifera* leaves, fruits and roots possess antioxidant properties and antibacterial activities. *BMC Complement Altern Med* 12: 175.
- [4] Parihar P, Shetty R, Ghafourifar P, Parihar MS (2016) Increase in oxidative stress and mitochondrial impairment in hypothalamus of streptozotocin treated diabetic rat: Antioxidative effect of *Withania somnifera*. *Cell Mol Biol (Noisy-le-grand)* 62: 73-83.
- [5] Choudhary B, Shetty A, Langade DG (2015) Efficacy of Ashwagandha (*Withania somnifera* [L.] Dunal) in improving cardiorespiratory endurance in healthy athletic adults. *Ayu* 36: 63-68.
- [6] Halder B, Singh S, Thakur SS (2015) *Withania somnifera* root extract has potent cytotoxic effect against human malignant melanoma cells. *PLoS One* 10: e0137498.
- [7] Verma SK, Kumar A (2011) Therapeutic uses of *Withania somnifera* (ashwagandha) with a note on withanolides and its pharmacological actions. *Asian J Pharm Clin Res* 4: 1-4.
- [8] Shah N, Singh R, Sarangi U, Saxena N, Chaudhary A, Kaur G, Kaul SC, Wadhwa R (2015) Combinations of ashwagandha leaf extracts protect brain-derived cells against oxidative stress and induce differentiation. *PLoS One* 10: e0120554.
- [9] Al-Awthan YS, Hezabr SM, Al-Zubairi AM, Al-Hemiri FA (2014) Effects of aqueous extract of *Withania somnifera* on some liver biochemical and histopathological parameters in male guinea pigs. *Pak J Biol Sci* 17: 504-510.
- [10] Nema R, Jain P, Khare S, Pradhan A, Gupta A, Singh D (2012) Study of *Withania somnifera* with the spatial reference of phytochemical, FTIR and flavonoids quantification. *Int J Pharm Life Sci* 3: 1530-1532.
- [11] Kumar V, Dey A, Hadimani MB, Marcović T, Emerald M (2015) Chemistry and pharmacology of *Withania somnifera*: An update. *Tang (Humanitas Medicine)* 5: e1.
- [12] Misra L, Mishra P, Pandey A, Sangwan RS, Sangwan NS, Tuli R (2008) Withanolides from *Withania somnifera* roots. *Phytochemistry* 69: 1000-1004.
- [13] Lala P, Misra L, Sangwana RS, Tuli R (2006) New withanolides from fresh berries of *Withania somnifera* Z. *Naturforsch* 61b: 1143-1147.
- [14] Zhao J, Nakamura N, Hattori M, Kuboyama T, Tohda C, Komatsu K (2002) Withanolide derivatives from the roots of *Withania somnifera* and their neurite outgrowth activities. *Chem Pharm Bull* 50: 760-765.
- [15] Budhiraja RD, Krishan P, Sudhir S (2000) Biological activity of withanolides. *J Sci Ind Res* 59: 904-911.

- [16] Baitharu I, Jain V, Deep SN, Shroff S, Sahu JK, Naik PK, Ilavazhagan G (2014) Withanolide A prevents neurodegeneration by modulating hippocampal glutathione biosynthesis during hypoxia. *PLoS One* 9: e105311.
- [17] Sangwan NS, Sabir F, Mishra S, Bansal S, Sangwan RS (2014) Withanolides from *Withania somnifera* Dunal: Development of cellular technology and their production. *Recent Pat Biotechnol* 8: 25-35.
- [18] Ku SK, Bae JS (2014) Antiplatelet, anticoagulant, and profibrinolytic activities of withaferin A. *Vascul Pharmacol* 60: 120-126.
- [19] Gao S, Li H, Zhou XQ, You JB, Tu DN, Xia G, Jiang JX, Xin C (2015) Withaferin A attenuates lipopolysaccharide-induced acute lung injury in neonatal rats. *Cell Mol Biol (Noisy-le-grand)* 61: 102-106.
- [20] Stenger VJ (1999) Bioenergetic fields. *Sci Rev Alternative Med* 3.
- [21] Rogers, M (1989) "Nursing: A Science of Unitary Human Beings." In J. P. Riehl-Sisca (ed.) *Conceptual Models for Nursing Practice*. 3<sup>rd</sup> Edn. Norwalk: Appleton & Lange.
- [22] Rubik B (2002) The biofield hypothesis: Its biophysical basis and role in medicine. *J Altern Complement Med* 8: 703-717.
- [23] Nelson LA, Schwartz GE (2005) Human biofield and intention detection: Individual differences. *J Altern Complement Med* 11: 93-101.
- [24] Nemeth L (2008) Energy and biofield therapies in practice. *Beginnings* 28: 4-5.
- [25] Koithan M (2009) Introducing complementary and alternative therapies. *J Nurse Pract* 5: 18-20.
- [26] Trivedi MK, Branton A, Trivedi D, Nayak G, Gangwar M, Jana S (2015) Bacterial identification using 16S rDNA gene sequencing and antibiogram analysis on biofield treated *Pseudomonas fluorescens*. *Clin Med Biochemistry Open Access* 1: 101.
- [27] Trivedi MK, Patil S, Shettigar H, Bairwa K, Jana S (2015) Effect of biofield treatment on phenotypic and genotypic characteristic of *Providencia rettgeri*. *Mol Biol* 4: 129.
- [28] Trivedi MK, Patil S, Shettigar H, Mondal SC, Jana S (2015) The potential impact of biofield treatment on human brain tumor cells: A time-lapse video microscopy. *J Integr Oncol* 4: 141.
- [29] Trivedi MK, Branton A, Trivedi D, Nayak G, Gangwar M, Jana S (2015) Antibiogram, biochemical reactions, and genotypic pattern of biofield treated *Pseudomonas aeruginosa*. *J Trop Dis* 4: 181.
- [30] Trivedi MK, Branton A, Trivedi D, Nayak G, Mondal SC, Jana S (2015) Biochemical differentiation and molecular characterization of biofield treated *Vibrio parahaemolyticus*. *American Journal of Clinical and Experimental Medicine* 3: 260-267.
- [31] Trivedi MK, Patil S, Shettigar H, Bairwa K, Jana S (2015) Phenotypic and biotypic characterization of *Klebsiella oxytoca*: An impact of biofield treatment. *J Microb Biochem Technol* 7: 203-206.
- [32] Trivedi MK, Patil S, Shettigar H, Gangwar M, Jana S (2015) An effect of biofield treatment on multidrug-resistant *Burkholderia cepacia*: A multihost pathogen. *J Trop Dis* 3: 167.
- [33] Trivedi MK, Branton A, Trivedi D, Shettigar H, Nayak G, Mondal SC, Jana S (2015) Antibiogram, biochemical reactions and genotyping characterization of biofield treated *Staphylococcus aureus*. *American Journal of BioScience* 3: 212-220.
- [34] Trivedi MK, Tallapragada RM (2008) A transcendental to changing metal powder characteristics. *Metal Powder Report* 63: 22-38, 31.
- [35] Dabhade VV, Tallapragada RMR, Trivedi MK (2009) Effect of external energy on the atomic, crystalline, and powder characteristics of antimony and bismuth powders. *Bull Mater Sci* 32: 471-479.
- [36] Trivedi MK, Patil S, Tallapragada RM (2013) Effect of bio field treatment on the physical and thermal characteristics of vanadium pentoxide powders. *J Material Sci Eng S* 11: 001.
- [37] Trivedi MK, Branton A, Trivedi D, Nayak G, Panda P, Jana S (2016) 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). *American Journal of Chemical Engineering* 4: 78-86.
- [38] Trivedi MK, Branton A, Trivedi D, Nayak G, Singh R, Jana S (2015) Evaluation of physical, thermal and spectroscopic properties of biofield treated *p*-hydroxyacetophenone. *Nat Prod Chem Res* 3: 190.
- [39] Trivedi MK, Branton A, Trivedi D, Nayak G, Mishra RK, Jana S (2015) Characterization of physical, thermal and spectral properties of biofield treated 2-aminopyridine. *Science Journal of Analytical Chemistry* 3: 127-134.
- [40] Trivedi MK, Mohan R, Branton A, Trivedi D, Nayak G, Latiyal O, Jana S (2015) Evaluation of biofield energy treatment on physical and thermal characteristics of selenium powder. *Journal of Food and Nutrition Sciences* 3: 223-228.
- [41] Trivedi MK, Nayak G, Patil S, Tallapragada RM, Jana S, Mishra RK (2015) Bio-field treatment: An effective strategy to improve the quality of beef extract and meat infusion powder. *J Nutr Food Sci* 5: 389.
- [42] Trivedi MK, Branton A, Trivedi D, Nayak G, Saikia G, Jana S (2015) Physical and structural characterization of biofield treated imidazole derivatives. *Nat Prod Chem Res* 3: 187.
- [43] Trivedi MK, Tallapragada RM, Branton A, Trivedi D, Nayak G, Mishra RK, Jana S (2015) Characterization of physical, spectral and thermal properties of biofield treated 1, 2, 4-Triazole. *J Mol Pharm Org Process Res* 3: 128.
- [44] Trivedi MK, Branton A, Trivedi D, Nayak G, Gangwar M, Jana S (2015) Agronomic characteristics, growth analysis, and yield response of biofield treated mustard, cowpea, horse gram, and groundnuts. *International Journal of Genetics and Genomics* 3: 74-80.
- [45] Trivedi MK, Branton A, Trivedi D, Nayak G, Mondal SC, Jana S (2015) Evaluation of plant growth, yield and yield attributes of biofield energy treated mustard (*Brassica juncea*) and chick pea (*Cicer arietinum*) seeds. *Agriculture, Forestry and Fisheries* 4: 291-295.

- [46] Devkar ST, Kandhare AD, Sloley BD, Jagtap SD, Lin J, Tam YK, Katyare SS, Bodhankar SL, Hegde MV (2015) Evaluation of the bioavailability of major withanolides of *Withania somnifera* using an *in vitro* absorption model system. *J Adv Pharm Technol Res* 6: 159-164.
- [47] Verma SK, Kumar A (2011) Therapeutic uses of *Withania somnifera* (ashwagandha) with a note on withanolides and its pharmacological actions. *Asian J Pharm Clin Res* 4: 1-4.
- [48] Cherson R (2009) Bioavailability, bioequivalence, and drug selection. In: Makoid CM, Vuchetich PJ, Banakar UV (eds) *Basic pharmacokinetics* (1<sup>st</sup> edn) Pharmaceutical Press, London.
- [49] Blagden N, de Matas M, Gavan PT, York P (2007) Crystal engineering of active pharmaceutical ingredients to improve solubility and dissolution rates. *Adv Drug Deliv Rev* 59: 617-630.
- [50] Trivedi MK, Mohan TRR (2016) Biofield energy signals, energy transmission and neutrinos. *American Journal of Modern Physics* 5: 172-176.
- [51] Khadka P, Ro J, Kim H, Kim I, Kim JT, Kim H, Cho JM, Yun G, Lee J (2014) Pharmaceutical particle technologies: An approach to improve drug solubility, dissolution and bioavailability. *Asian J Pharm Sci* 9: 304-316.
- [52] Buckton G, Beezer AE (1992) The relationship between particle size and solubility. *Int J Pharmaceutics* 82: R7-R10.
- [53] Reichenbacher M, Popp J (2012) Challenges in Molecular structure determination, DOI 10.1007/978-3-642-24390-5\_2, Springer-Verlag Berlin Heidelberg.
- [54] Stuart BH (2004) *Infrared spectroscopy: Fundamentals and applications in Analytical Techniques in the Sciences*. John Wiley & Sons Ltd., Chichester, UK.
- [55] Joshi P, Misra L, Siddique AA, Srivastava M, Kumar S, Darokar MP (2014) Epoxide group relationship with cytotoxicity in withanolide derivatives from *Withania somnifera*. *Steroids* 79: 19-27.
- [56] Misra L, Lal P, Chaurasia ND, Sangwan RS, Sinha S, Tuli R (2008) Selective reactivity of 2-mercaptoethanol with 5 $\beta$ , 6 $\beta$ -epoxide in steroids from *Withania somnifera*. *Steroids* 73: 245-251.
- [57] Gu M, Yu Y, Gunaherath GMKB, Leslie Gunatilaka AA, Li D, Sun D (2014) Structure-activity relationship (SAR) of withanolides to inhibit *Hsp90* for its activity in pancreatic cancer cells. *Invest New Drugs* 32: 68-74.
- [58] Ramachandran A, Kumar MS (2014) FT-IR, UV and antimicrobial activity *Withania somnifera* and *Withania obtusifolia*. *Int J Pharm Bio Sci* 5: (B) 111-117.
- [59] Hesse M, Meier H, Zeeh B (1997) *Spectroscopic methods in organic chemistry*, Georg Thieme Verlag Stuttgart, New York.
- [60] Srivastava A, Alam S, Shahbaaz S, Tiwari M, Mittal A, Chauhan S (2014) Formulation and evaluation of antiacne cream containing *Withania somnifera*. *J Pharm Sci Inv* 3: 348-352.
- [61] Mitra D, Francis S, Varshney L (2004) Calorimetry thermal investigations on  $\gamma$  radiation processed natural medicinal products (ashwagandha, amla and hartiki). *J Therm Anal Cal* 78: 821-829.