some alternate strategies, which could improve the stability of BHT by altering their physical, thermal or structural and bonding properties.

Nowadays, biofield energy treatment is known to alter physicochemical properties of various organic and metallic compounds [16-19]. The biofield energy healing therapies are considered as complementary and alternative medicine (CAM) by National Center for Complementary and Alternative Medicine (NCCAM)/National Institute of Health (NIH) and are based on alteration in putative energy fields and consciousness [20,21]. The biofield energy is related to the energy associated with the human body that depends upon the physiological and mental health of the human. This energy can be exchanged with the environment through natural exchange process [22]. A human has the ability to harness the energy from environment or universe and can transmit in any living or non-living object(s) around the Universe. The objects always receive the energy and responding in a useful way, this process is known as biofield energy treatment. Moreover, the biofield therapies are reported for the reduction in pain, anxiety and tension [23]. Mr. Trivedi is well known to possess the unique biofield energy treatment (The Trivedi Effect) which is reported to alter the properties such as growth and yield of plants in the field of agriculture [24,25]. The effect was also reported on phenotypic characters of microorganisms in the field of microbiology [26,27]. Hence, the present study was designed to analyse the impact of biofield energy treatment on various physicochemical properties of BHT using XRD, DSC, TGA/DTG, FT-IR and UV-Vis spectroscopic techniques.

Materials and Methods

Material procurement

Butylated hydroxytoluene (BHT) was procured from S D Fine Chemicals Pvt. Ltd., India. After procurement, the BHT sample was divided into two parts; coded as control and treated, and stored as per manufacturer's guidelines.

Treatment modality

The treated part was subjected to Mr. Trivedi's biofield energy treatment. For this, the treated sample was handed over to Mr. Trivedi in sealed pack for biofield energy treatment under standard laboratory conditions. Mr. Trivedi provided the treatment to the treated group through his energy transmission process. The biofield treated sample was returned in the same sealed condition for further characterization using XRD, DSC, TGA, FT-IR and UV-Vis spectroscopic techniques.

X-ray diffraction (XRD) study

X-ray powder diffractogram of control and treated samples were obtained on Phillips, Holland PW 1710 X-ray diffractometer system. The X-ray generator was equipped with a copper anode with nickel filter operating at 35 kV and 20 mA. The wavelength of radiation used by the XRD system was 1.54056 Å. The data were collected from the 2θ range of 10°-99.99° with a step size of 0.02° and a counting time of 0.5 seconds per step.

The crystallite (G) was calculated from the Scherrer equation with the method based on the width of the diffraction patterns obtained in the X-ray reflected crystalline region.

 $G = k\lambda/(bCos\theta)$

Where, k is the equipment constant (0.94), λ is the X-ray wavelength (0.154 nm), b in radians is the full-width at half of the peak and θ the

corresponding Bragg angle. Other parameters *viz*. lattice parameter and unit cell volume, were calculated using PowderX software. Further, these parameters were used to calculate the molecular weight and density of the control and treated sample.

Percent change in crystallite size was calculated using the following equation:

Percent change in crystallite size = $[(G_t-G_c)/G_c] \times 100$

Here, G_c and G_t denotes the crystallite size of control and treated powder samples, respectively. Similarly the percent change in lattice parameter, unit cell volume, molecular weight, and density was calculated to analyse the impact of biofield treatment on crystal parameters of treated sample as compared to the control.

The molecular weight of atom was calculated using following equation:

Molecular weight = number of electrons \times weight of an electron + number of neutrons \times weight of a neutron + number of protons \times weight of a proton.

The weight of all atoms in a molecule was multiplied by the Avogadro number (6.023 \times 10^{23}) to obtain the molecular weight in g/ Mol.

Differential scanning calorimetry (DSC) study

DSC analysis of control and treated sample was carried out using Perkin Elmer/Pyris-1. Each sample was accurately weighed and hermetically sealed in aluminium pans and heated at a rate of 10° C/ min under air atmosphere (5 mL/min). The thermogram was collected over the temperature range of 50° C to 250° C. An empty pan sealed with cover pan was used as a reference sample. From DCS curve, the melting temperature and latent heat of fusion were obtained.

The percent change in latent heat of fusion was obtained using following equations to observe the difference in thermal properties of treated BHT sample as compared to the control:

% change in latent heat of fusion =
$$\frac{\left[\Delta H_{\text{Treated}} - \Delta H_{\text{Control}}\right]}{\Delta H_{\text{Control}}} \times 100$$

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

Thermogravimetric analysis/Derivative Thermogravimetry (TGA/DTG)

The effect of temperature on the stability of the control and treated sample of BHT was analysed using Mettler Toledo simultaneous thermogravimetric analyser (TGA/DTG). The samples were heated from room temperature to 350°C with a heating rate of 5°C/min under air atmosphere. From TGA/DTG curve, the onset temperature T_{onset} (temperature at which sample start losing weight) and T_{max} (maximum thermal degradation temperature) were recorded.

Fourier transform-infrared (FT-IR) spectroscopic characterization

For FT-IR characterization, the treated sample was divided into two groups named T1 and T2. The samples were crushed into fine powder for analysis. The powdered sample was mixed in spectroscopic grade KBr in an agate mortar and pressed into pellets with a hydraulic press. FT-IR spectra were recorded on Shimadzu's Fourier transform infrared spectrometer (Japan). FT-IR spectra are generated by the absorption of Citation: Trivedi MK, Branton A, Trivedi D, Nayak G, Singh R, et al. (2015) Physicochemical and Spectroscopic Characterization of Biofield Treated Butylated Hydroxytoluene. J Food Ind Microbiol 1: 101.

electromagnetic radiation in the frequency range 4000-400 cm⁻¹. With the help of FT-IR analysis, the impact of biofield treatment on bond strength, rigidity and stability of BHT compound can be analysed [28].

UV-Vis spectroscopic analysis

For UV-Vis spectroscopic analysis, the treated sample was divided into two groups, served as T1 and T2. The UV-Vis spectral analysis was measured using Shimadzu UV-2400 PC series spectrophotometer. The spectrum was recorded with 1 cm quartz cell having a slit width of 2.0 nm over a wavelength range of 200-400 nm. With UV-Vis spectroscopy, it is possible to investigate electron transfers between orbitals or bands of atoms, ions and molecules from the ground state to the first excited state [29].

Results and Discussion

X-ray diffraction (XRD)

The X-ray powder diffractograms of control and treated samples showing Bragg angle (2 θ) on x-axis and intensity of the peaks on y-axis are presented in Figure 2. The XRD diffractograms showed a series of sharp peaks in the regions of 10°<2 θ >40°, which depicted that both samples had high crystallinity and long range order of molecules. The sharp peaks on the diffractograms of the control and treated sample confirm the crystalline nature of BHT [30]. The XRD pattern indicated the orthorhombic crystal structure in the control and treated BHT samples. The crystal structure parameters were computed using PowderX software such as lattice parameter, unit cell volume, density, and molecular weight. The results are presented in Table 1. The data showed that in the control sample, the lattice parameters were found as a=15.46, b=10.37, and c=8.93 Å. While, in treated sample, the lattice parameters were found as a=15.59, b=10.56 and c=8.71 Å. It showed that the lattice parameters 'a' and 'b' were increased by 0.84% and



Parameter	Control	Treated	Percent change
Lattice parameter			
a (Å)	15.46	15.59	0.84
b (Å)	10.37	10.56	1.83
c (Å)	8.93	8.71	-2.46
Unit cell volume (×10 ⁻²³ cm ³)	143.208	143.330	0.090
Density (w/cm ³)	1.029	1.028	-0.10
Molecular weight (g/mol)	222.752	222.941	0.08
Crystallite size (nm)	70.16	59.77	-14.81

Table 1: X-ray diffraction analysis of control and treated butylated hydroxytoluene.

1.83%, respectively. However, the lattice parameter 'c' was decreased by 2.46% in the treated sample as compared to the control. Similarly, the unit cell volume and molecular weight was slightly increased by 0.09% and 0.08%, respectively; whereas, density was decreased by 0.10% in the treated sample as compared to the control. The increase in unit cell volume and change in lattice parameters indicated the presence of internal strain in the treated BHT powder. Besides, the crystallite size computed using Scherrer formula was found as 70.16 nm in control and it was reduced to 59.77 nm in the treated BHT sample. It suggested that crystallite size of the treated sample was significantly reduced by 14.81% as compared to the control. It is reported that the energy produced by mechanical milling had reduced the crystallite size and induced lattice strain in the crystal structure [31]. Thus, it is assumed that biofield energy treatment might induce the energy milling in BHT sample, and that might be responsible for a decrease in crystallite size of treated sample. Recently, our group reported that biofield treatment had reduced the crystallite size in magnesium powder [19]. Moreover, it was reported that crystallite size and surface area are inversely related to each other [32]. The BHT had a poor solubility profile in water that limits its application in pharmaceutical preparations [7]. Hence, the decreased crystallite size of treated BHT might result in increased surface area, and that can play an important role in improving solubility. Therefore, the treated BHT sample may be used in food and pharmaceutical industry with improved solubility profile.

DSC analysis

This technique is based on the principle that as the BHT sample undergoes any phase transition (e.g. solid to liquid); the alteration was observed in the amount of heat flowing to the sample as compared to the reference. The thermograms for control and treated sample of BHT are presented in Figure 3 that showed the phase transition temperature and the amount of heat involved in that process. The control sample exhibited a sharp endothermic peak at 71.6°C, whereas the treated sample showed a sharp peak at 72.25°C. The peaks are due to melting of control and treated samples, and the sharpness of peaks confirms the crystalline nature of BHT sample. The result suggests a slight change in melting temperature of treated sample as compared to the control. The thermograms also showed that the latent heat of fusion (Δ H) was increased from 75.94 J/g (control) to 96.23 J/g in treated BHT sample. It indicated that ΔH was significantly increased by 26.72% in the treated sample as compared to the control. It is presumed that biofield energy might increase the potential energy stored in the molecules of treated BHT sample. Hence, the treated sample needs more energy in the form of ΔH to undergo the process of melting. Previously, our group reported that biofield treatment has altered the latent heat of fusion in indole, thymol and menthol compounds [17,33].