PAPER • OPEN ACCESS

Spectroscopic investigation of tau protein conformational changes by static magnetic field exposure

View the article online for updates and enhancements.

Journal of Physics Communications

PAPER

Spectroscopic investigation of tau protein conformational changes by static magnetic field exposure

Saqer M Darwish[1],* and Imtiaz M Darwish[2]

[1] Biophysics lab, Physics Department, Al-Quds University, Abu-Dies Jerusalem, Palestine
[2] Chemistry Department. Birzeit University, Birzeit, Ramallah, Palestine
* Author to whom any correspondence should be addressed.

E-mail: sdarwish@staff.alquds.edu, ikhalid@birzeit.edu and sdarwish@science.alquds.edu

Abstract

Electromagnetic fields taint the molecular environment of proteins and induce changes in the central nervous system. This research applied Fourier transform infrared spectroscopic analysis to investigate the effects of static magnetic fields on tau protein in neurological disorders. It explores the conformational changes of tau protein and highlights its' potential application as a pathological biomarker for early detection and therapeutic interventions. The results indicate that tau protein is susceptible to magnetic field exposure in the amide B, fingerprint, and amide regions (IV-VI). Changes in peak positions and band intensities were identified and delineated as the outcome effect of magnetic forces on molecular vibrations. Magnetic forces may affect the microtubule structure of the tau protein, leading to protein aggregation. These results indicate the potential application of FTIR spectroscopy for the early detection and classification of degenerative diseases through spectrum analysis. Different magnetic fields can be used as spontaneous therapeutic procedures to induce changes in the molecular environment of proteins.

1. Introduction

The human brain contains billions of interconnected neurons and has different parts to carry out various functions effectively. Its contents are protected by the skull, which forms a closed environment that provides suitable conditions and eliminates any external interference in neural activities. The human brain functions as a highly integrated complex dynamic network that keeps its spatially distributed regions sharing information [1]. Any local disturbance that causes a functional change in one part affects the others; therefore, all parts are connected to continue coordination without any external interference.

Electronic devices and industrial technology are producing radiation pollution at an accelerating rate. Electromagnetic fields are becoming more pervasive to the human brain through the overwhelming number of communication devices. Usually, sensitive protein functions and neurological activities are performed in an isolated environment without external influences. There is a need for more scientific investigations directed at the molecular pollution caused by electromagnetic fields and other forms of radiation.

Recent experimental evidence has shown that exposure to a transcranial static magnetic field (TSMF) can affect specific functions within the human brain. Exposure to TSMF reduces motor cortical excitability and regulates excitation balance towards inhibition. Besides, exposure to TSMF can lower the discomfort caused by the high light intensity in humans [2, 3].

Neuromodulation of the human brain through exposure to magnetic fields is conforming as a simple means for treating various neurological and neuropsychiatric disorders. Several experiments showed that TSMF had reduced the excitability of the motor cortex for a reasonable amount of time. These results encourage the use of small static magnets to modulate cerebral excitability in a non-intrusive, painless, and reversible way. However, the mechanisms by which static magnetic fields modulate cortical activity are considerably unknown. During
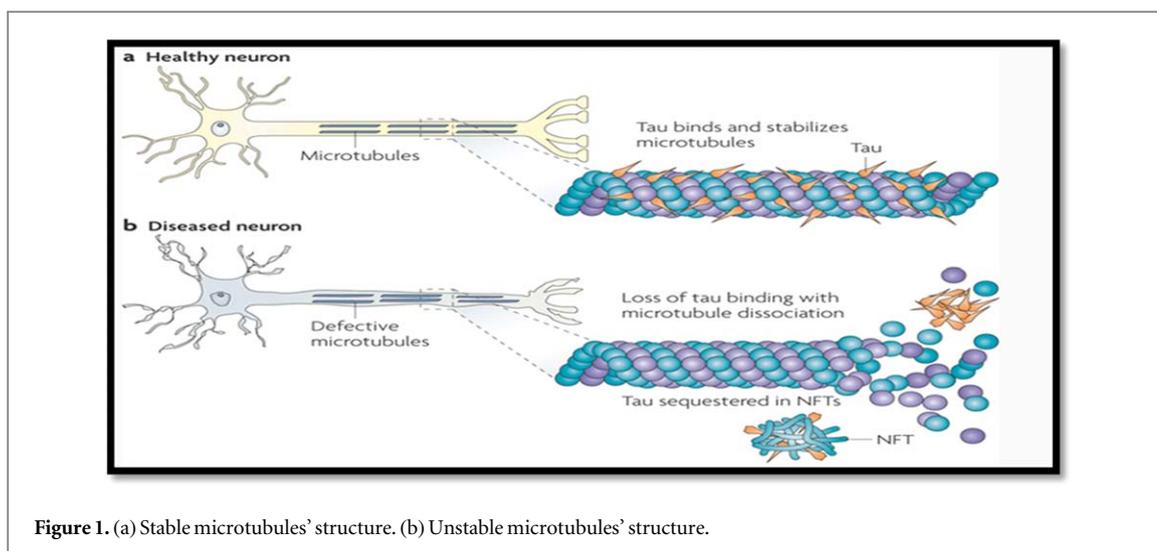
Figure 1. (a) Stable microtubules' structure. (b) Unstable microtubules' structure.

the last decade, several studies indicated that moderate TSMF in the range between 1 mT and 1 T can affect human cortical excitability [4]. TSMF has been used as a noninvasive brain stimulation technique, but more investigations are needed to understand these mechanisms [5–7]. Another area of scientific concern is whether exposure to static magnetic fields causes DNA damage or not. Past studies have confirmed that a static magnetic field alone has no such effect on the fundamental properties of cell growth and survival under standard culture conditions.

However, there is an indication that the frequency of micronucleus formation changes substantially when specific treatments such as X-irradiation and mitomycin C are used during exposure to a strong static magnetic field. Several studies have suggested that a strong static magnetic field may influence ion transport and gene expression. Other studies have found that a strong magnetic field can cause spatial orientation phenomena in cell culture [8]. Moreover, protein misfolding and aggregation are becoming fingerprints of neurodegenerative diseases, including Alzheimer's, Huntington's, Parkinson's, prion diseases, and amyotrophic lateral sclerosis [9]. Protein misfolding and aggregation are natural phenomena that disrupt cell function through many mechanisms that are yet to be fully explained [10, 11].

In 2002, The International Agency for Research on Cancer (IARC) revealed that extremely low frequency (ELF) magnetic fields are 'possibly carcinogenic to humans' [12–15]. Most of the neural activities in the brain are processed through electrical impulses. Consequently, it is possible that exposure to a static magnetic field (SMF) can yield physiological and biochemical alterations. However, there is a lack of unanimity regarding the effects of SMF on different proteins; it appears that SMF induces physiological changes in exposed proteins. These changes can either improve biological tasks or cause dysfunction depending on the amount of absorbed energy by the proteins [16–20].

Tau is a microtubule-associated protein that has an essential role in axonal stabilization, neuronal development, and neuronal polarity [21]. It composes cytoskeletons which are crucial in maintaining spine development and morphology, brain cognitive behavior, consciousness, and memory storage [22]. These skeletons form a tube-shaped where nutrients and other substances are carried through into the neurons. Tau proteins are intrinsically disordered due to their unstable conformation and high pliability [23]. The role of tau protein is to maintain healthy neurons through reversible polymerization of microtubules formations to maintain neural growth and neural polarity [24]. Figure 1 illustrates the function of Tau protein in keeping neurons healthy [25].

Tau protein determinations and quantifications are essential to the diagnosis of most degenerative diseases. Early detections of tau protein changes based on biomarkers may allow earlier diagnosis for most infectious and autoimmune diseases [26]. Researchers are becoming more convinced that Alzheimer's disease (AD) may result from a combination of abnormal tau and beta-amyloid proteins associated with other age-related diseases such as cerebrovascular and Lewy body [27]. Analysis of several brain tissues from AD patients confirmed two different abnormal formations of extra neural amyloid plaques and intraneural tangles containing fibrillary structure mainly composed of microtubule Tau protein [27–31].

It seems that neuropathologic diagnosis of AD cases correlates to the formation of both filamentous tau protein and cored neuritic plaques [32]. The tau filaments are called 'paired helical filaments' (PHFs). They reveal reparative patterns as viewed through electron microscopy showing two tiny filaments twisted around one another, forming periodic structures with a 65–80 nm crossover distance. On the other hand**,** neuritic plaques

are composed of activated microglia and reactive astrocytes entangled with neuritic elements in the plaque periphery [27]. In other words, the abnormal aggregation of tau protein into insoluble paired helical filaments (PHFs) is one of the indications of AD [33]. It appears that intracellular inclusions of fibrillar forms of tau protein with β-sheet structure assemble in specific brain regions, causing memory loss. Besides, pathological studies using animals and humans have shown that tau oligomer formation contributes to neuronal loss [23]. Microtubule-associated protein tau belongs to the expanding group of natively unfolded proteins or intrinsically disordered proteins (IDPs), which can display novel features in protein chemistry.

Furthermore, histological analysis and tau positron emission tomography (PET) imaging studies have disclosed that cognitive impairment correlates with tau presence and neuronal loss in the human brain and its associations with aging and AD [34]. It suggests that tau oligomers formation and grey matter loss lead to cognitive deficits by different mechanisms, suggesting implications for future therapeutic trials targeting tau pathology [35]. Another analysis suggests that aggregation of hyperphosphorylated forms of tau in the neural soma forming neurofibrillary tangles may constitute the leading cause of AD [36]. Heiko and Eva Braak have shown that tau deposits of neurofibrillary tangles follow a particular pathway in their proliferating from the trans entorhinal cortex to neocortical association areas and finally to secondary and primary cortical areas. They suggested six disease stages where the first two stages are asymptomatic, the following two stages show some cognitive ability loss, and the last two stages are associated with dementia [37].

Previous studies have shown that the vibration modes of the amide bands exhibit high sensitivity when exposed to ELF-electromagnetic fields [38, 39]. Protein conformational changes are observed because of the effects on vibrational modes of the amide bands in proteins in the IR region of the spectrum [40]. Fourier-transform infrared (FTIR) spectroscopy is a noninvasive technique to detect changes in the secondary structure of proteins and has been widely used to investigate protein oligomerization, aggregation, and fibril formation [10, 11]. A considerable amount of research linked protein conformational changes to the molecular vibration of the absorption bands of the secondary structure in the studied proteins. These changes can be detected through the spectrum variations in the mid-infrared range (400–4000 cm^{-1}). FTIR is a sensitive diagnostic tool to differentiate abnormalities in biological cells. The biochemical changes in cells often correspond to changes in the fingerprint regions of the proteins.

The present study will investigate the impact of exposing tau protein to SMF by FTIR spectroscopy. The experimental setup aims to imitate neuronal proteins' exposure to SMF, similar to what happens in the brain by passing neuron signals. A considerable amount of scientific research has revealed a strong connection between band intensities and their peak positions of the IR spectra to the secondary structure changes of the studied proteins [41–43].

Studies of the effects of SMF on different proteins could open new perspectives in health research, especially in the applicability of immunosensor development in clinical diagnosis. Several studies have used biological samples obtained from patients with neurodivergent conditions to confirm the application of immune sensors through the time-dependent phase angle shift technique Single Frequency Analysis (SFA), which is based on the phase angle shift from antibody-antigen interactions [44].

Early detection of tau oligomers formation through absorbed infrared energy can be used as a sensor to detect certain diseases' early stages accurately. The absorbance spectrum in the Mid-infrared region for a tissue or blood sample is considered a unique way to represent the contents of the sample. The unknown sample is easily identified by comparing the obtained fingerprint spectrum to a library of known spectra. The analysis of the measured spectra can provide insights into physiological and pathological changes that have taken place at the molecular level.

2. Materials and methods

2.1. Preparation of stock solutions

Tau protein contains six recombinant tau proteins expressed in *E. coli*, with molecular weights of 36.8, 39.7, 40.0, 42.6, 42.9, and 45.9 kDa. The proteins are expressed without histidine tags. The purity of each protein is >90% (SDS-PAGE). The six Tau isoforms differ from one another by the number (3 or 4) of microtubule-binding repeats (R) of 31–32 amino acids each and the number (0, 1, or 2) of amino-terminal inserts (N) of 29 amino acids each [45]. Tau protein is supplied as a solution in 125 mM Tris-HCl, pH 6.8, with 4% SDS, 10% 2-mercaptoethanol, 20% glycerol, and 0.004% bromophenol blue. Each sample of 50 ml of the tau protein ladder contains 0.25 mg of each of the six isoforms.

The Tau protein samples were obtained from Sigma Aldrich Chemical Company. The samples were kept at −20 °C without any further purification prior to their use. Samples solutions were incubated for one hour at room temperature, then the samples were prepared for measurement. 50 ml of each aqueous sample was

positioned on a silicon window plate and was left to dry completely at room temperature prior to taking spectroscopic measurements.

2.2. FTIR spectroscopic measurements

Bruker IFS 66/S spectrophotometer equipped with a liquid nitrogen-cooled Mercury-Cadmium-Telluride (MCT) detector and KBr beam splitter was used to scan the FTIR measurements. The spectrophotometer was continuously purged with dry air to reduce the noise signal. Each spectrum was obtained by taking an average of 60 repeated scans to maintain measurement accuracy. The FTIR measurements were obtained at a spectral resolution of 4 cm^{-1}, and the aperture setting was held at 8 mm, which gives the best signal-to-noise ratio. OPUS software is used to accurately make the baseline corrections, the second derivatives of the spectra, and peak position identifications. The Fourier self-deconvolution (FSD) process decomposes the major absorption bands into the original constituents of each individual peak, which correspond to the vibrational modes of molecules in the protein's secondary structure. All FSD processes were consistently repeated six times. The FTIR spectra of tau protein were scanned in the featured region of (4000–400) cm^{-1}. The background's absorption spectrum was consistently subtracted from the sample's spectrum from each of the FTIR spectra. The net effect of the static magnetic field on the tau protein, represented by the difference spectra, was calculated by subtracting the sample's spectrum before exposure to the magnetic field from the sample's spectrum after exposure. As an accuracy check, the resulting difference spectra of the featureless region of the protein spectra (1800–2200) cm^{-1}, must always give zero difference. In addition, control samples with the same protein concentration difference spectra resulted in a flat line formation as expected.

2.3. Static magnetic field measurements

DC voltage regulated at 220 volts and connected to an electromagnetic coil was used to produce a static magnetic field. The prepared sample was placed as close as possible to the center of the coil at a fixed holder. This sample positioning ensures that the magnetic field is uniform and perpendicular to the sample's surface. The Tau sample experienced the following various magnetic fields: 0.0, 0.24, 0, 48, 0.6, 0,9, 0.45, 1.2, 1.8, 2.2 mT. Each sample was exposed to each magnetic field setting for 2.0 min. The value of each magnetic field was taken for each measurement using a magnetic field axial probe PHYWE Gauss meter.

3. Results

The experimental results were obtained by analysis of FTIR absorption spectra for tau protein over the entire mid-infrared region (4000–400) cm^{-1} as shown in figure 2(A). At the same time, the second derivative of the absorption spectrum is shown in figure 2(B).

The FTIR spectra were obtained for Tau Protein before and after the exposure to different static magnetic fields while keeping the sample fixed in its position all the time. This approach allows for precise comparison of band intensities and peak positions before and after exposing the protein to the different static magnetic fields.

The FSD absorbance spectra of the tau protein showed detailed structure for the significant absorption bands including, all amide bands and the fingerprint region. The absorption peaks in the amide I region (1600–1700) cm^{-1} correspond to the stretching vibrations of the C=O coupled with C-N stretching and C-C-N deformation mode [46].

Several peaks are shown in the amide II region (1480–1600) cm^{-1} due to the out-of-plane merger of N-H in-plane bending and C-N stretching vibration [47]. The absorption bands in the amide III region (1220–1320) cm^{-1} are composed of an in-plane mixture between the N–H bending and the C–N stretching with extra contributions from the C–H and the N–H deformation vibrations [47, 48]. The absorption bands in the amide IV region (625–770) cm^{-1} correspond to the vibration linked to a mixture of OCN bending vibrations coupled with out-of-plane N–H bending [49]. In the amide V region (640–800) cm^{-1}, the absorption bands are due to out-of-plane N-H bending, while the absorption bands in the VI region (537–606) cm^{-1} are due to out-of-plane C=O bending [50]. The absorption bands in the region (900–1220) cm^{-1} are assigned to C–O bending vibrations of saccharides (glucose, lactose, and glycerol) [51]. Furthermore, the absorption bands in the region (1360–1430) cm^{-1} are due to the vibrations of specific amino acid chains, while the absorption bands in the range of (1430–1480) cm^{-1} are caused by fatty acids, phospholipids, and triglycerides [52].

All the prominent seen peaks of the absorption bands before and after exposure to magnetic fields in the mid-infrared spectral range (400–4000) cm^{-1} are assigned according to the second derivative, and the FSD computations are listed in table 1. The FSD absorption spectra of tau protein show the main absorption bands, including the amide bands and the fingerprint regions.

The effect of exposing tau protein to a static magnetic field at various magnitudes (0.0, 0.24, 0.48, 0.6, 0.9, 1.2, 1.8, 2.2) mT is revealed by several FSD absorption spectra changes.
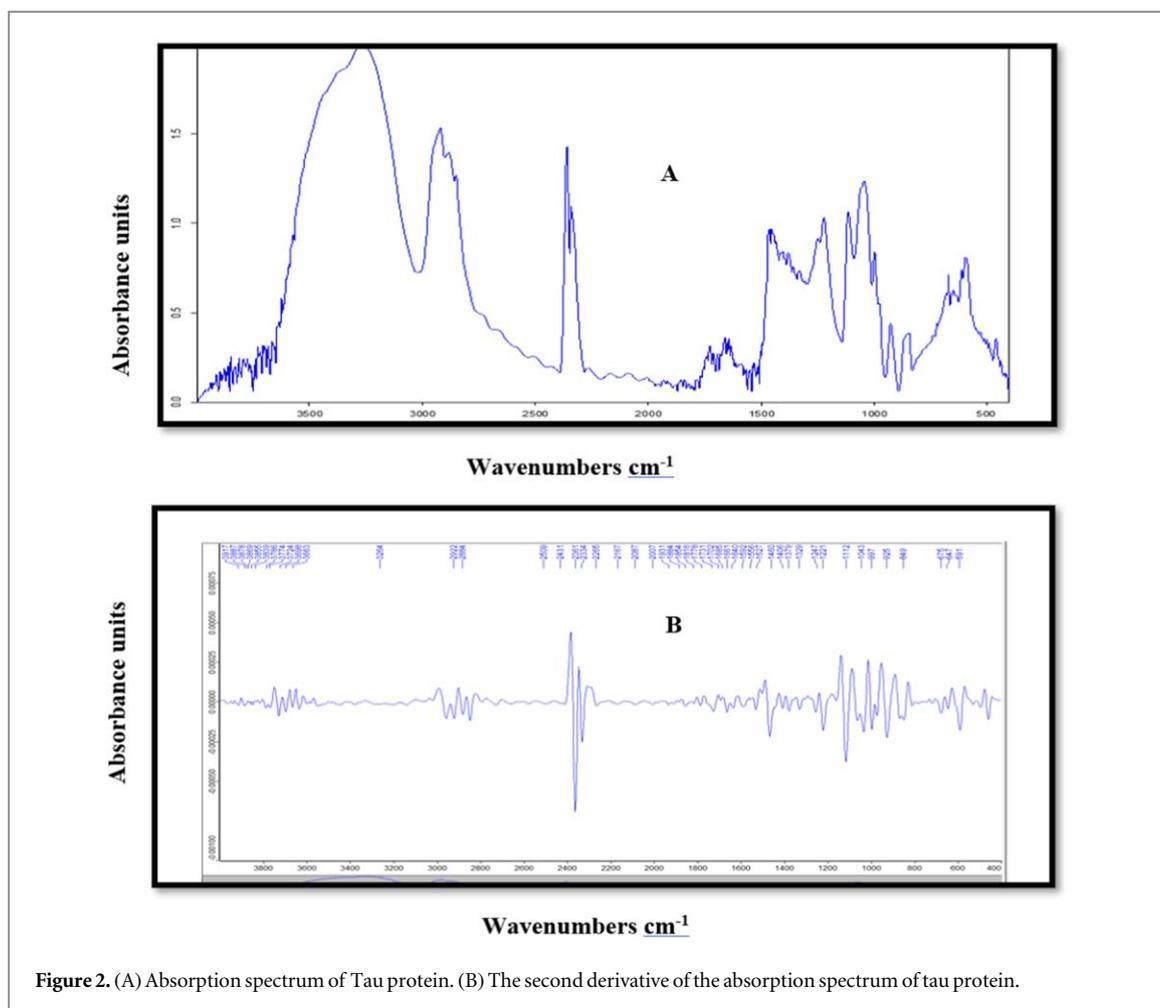
Figure 2. (A) Absorption spectrum of Tau protein. (B) The second derivative of the absorption spectrum of tau protein.

The spectra in figure 3 for the range of (500–1000) cm^{-1} show a remarkable increase in most bands' intensities due to the exposure to the static magnetic field.

Moreover, peak shifts with minor changes in the band structures were observed in some cases, indicating high sensitivity to the magnetic field. The peak at 583 cm^{-1} merged with the neighboring peak at 594 cm^{-1} after the magnetic field exposure. The bands at (595 and 612) cm^{-1} have tripled in their intensities, and the peak at 612 cm^{-1} shifted to 620 cm^{-1}. The peak at 632 cm^{-1} decreased in intensity and appeared as a shoulder with the neighboring band at 650 cm^{-1} as the magnetic field strength increased. The peak at 669 cm^{-1} did not change, while the peaks at 683, 704, and 719 have shown a small increase in their intensities. The peak at 737 cm^{-1} reduced its intensity sharply and disappeared, while the peak at 800 cm^{-1} increased its intensity suddenly after exposure to the magnetic field. The remaining peak' changes are listed in table 1. The FSD spectra in the ranges of (1000–1700) cm^{-1} and (1700–4000) cm^{-1} are shown in figure 4 and figure 5 respectively. All changes in band intensities and peak positions are listed in table 1 and table 2.

Consistently, the resultant effect of the magnetic field on the tau protein is determined by subtracting the tau protein's spectrum before exposure to the magnetic field from the spectrum after the exposure. The difference spectra for the range (500–1700) cm^{-1} is shown in figure 6(C), which reveals all the affected absorption bands by the exposure to the SMF.

The peaks of the difference spectrum, which represent the net changes in the molecular vibrations of the exposed tau protein, are listed in Table 1. The area under the bands above the zero line represents the net effect on the intensities of the absorption bands of tau protein due to its exposure to the magnetic field. For the amide regions (IV-VI), the (596 and 619) cm^{-1} peaks showed a strong effect upon magnetic field exposure on tau protein, while the peaks at (515, 670, 723, and 800) cm^{-1} reveal a moderate effect by the exposure. In addition, figure 6(C). reveals other noticeable peaks at the positions: (819, 832, 849, 951, 988, 1005, 1050, 1113, 1219, 1249, 1367, 1387, and 1425) cm^{-1}. These peaks correlate to the increase of bands' intensities because of the magnetic field effect on the tau protein.

Figure 7(C) shows the difference spectra for the region (1700–4000) cm^{-1}, which reveals the impact of the magnetic field on tau protein through the changes in the spectrum.

Table 1. Band positions (± 0.5 cm^{-1}) before and after tau exposure to SMF.

Amide Bands and Regions	Peak position at 0.0 mT	Peak position at 2.2 mT	Intensity change	Difference spectra peak position	Inverted peak position
Amide VI	512	513	increase	514	
	528	528	no change		
	544	544	no change		
	561	563	no change		
	583	—	decrease		583
	595	595	increase	596	
Amide V & IV	612	620	increase	619	
	632	636	decrease	640	610
	650	650	increase	654	632
	669	669	increase	669	
	683	683	Increase	683	
	704	704	increase	704	
	719	721	increase	723	
	737	737	decrease	746	
	761	761	increase	762	737
	780	780	increase	798	
	—	800	increase	800	
	809	819	increase	819	
	823	832	decrease	832	
	840	850	increase	849	826
	869	869	increase	866	
	885	890	increase	890	
900–1200	905	910	increase	909	
	927	927	increase	926	
	952	952	increase	951	
	977	977	decrease	964	
	998	998	decrease	988	977
				1005	998
	1023	1029	increase	1032	
	1043	1050	increase	1050	
	1063	1075	decrease	1077	
	1091	1095	increase	1093	1063
	1118	1115	increase	1113	
	1195	1198	increase	1197	
Amide III	1430	1427	increase	1425	
		1441	increase	1441	
	1450	1451	decrease		
	1468	1468	no change		1450
Amide II	1482	1482	no change		
	1500	1500	no change		
	1517	1517	no change		
	1532	1532	no change		
	1550	1550	no change		
	1568	1568	no change		
	1586	1586	no change		
Amide I	1600	1600	no change		
	1612	1612	no change		
	1630	1630	no change		
	1644	1644	no change		
	1662	1662	no change		
	1678	1678	no change		
	1694	1694	no change		

The main differences are represented by the appearance of the following major peaks at these positions (1708, 1736, 1757, 2338, 2356, 2849, 2918, and 2952) cm^{-1}. Again, the total area under the peaks corresponds to the enhancement of the bands' intensities upon exposure to SMF. Similarly, but to a lesser degree, the following bands at (2274, 2295, 2309, 2328, 2340, 2358, 2829, 2850, 2872, 2918, 2934, and 2960) cm^{-1} are also increased in their intensities. The data in table 2 reveals no other significant changes in band positions for all peaks in the range (1700–4000) cm^{-1}. The bands around 2200–2400 cm^{-1} indicate the possible presence of a C-N or a C-C triple bond [53]. The region 2820–2980 cm^{-1} (amide B) is associated with the C-H stretching vibrations of
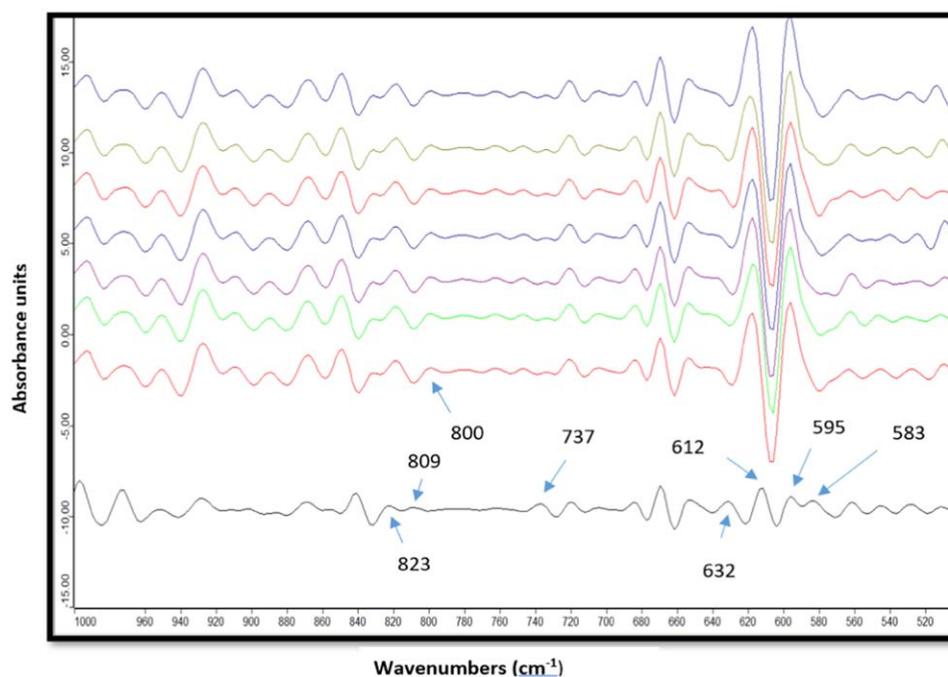
Figure 3. FSD spectra for tau protein exposed to SMF (0.0, 0.24, 0.48, 0.60, 0.90, 1.20, 1.8, 2.2) mT in order from bottom to up.
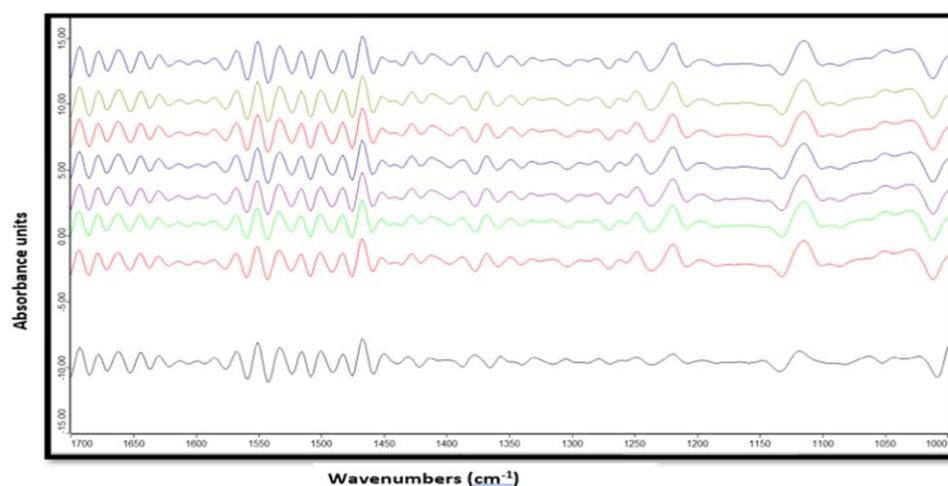

Figure 4. FSD spectra for tau Protein exposed to SMF (0.0, 0.24, 0.48, 0.60, 0.90, 1.20, 1.8, 2.2) mT in order from bottom to up.

lipids. The band at (2918 cm^{-1}) is due to asymmetric C-H stretching vibration, while the band at (2850 cm^{-1}) is due to symmetric C-H stretching vibration [54]. The bands at 2850 cm^{-1} and 2960 cm^{-1} are due to the symmetric CH$_2$ stretching mode and asymmetric stretching mode, respectively, in membrane lipids [55].

4. Discussion

The experimental results of exposing tau protein to SMF have shown changes in the measured absorption spectra. These changes are related to changes in the secondary structure of tau protein and are indicated by band position, band intensity, and bandwidth variations for most of the observed bands. The peaks' changes are usually linked to a shift in the absorbed frequency due to external effects on the vibrational bonds. The frequency of vibration of the normal mode excited by the absorbed light is expressed by equation (1).
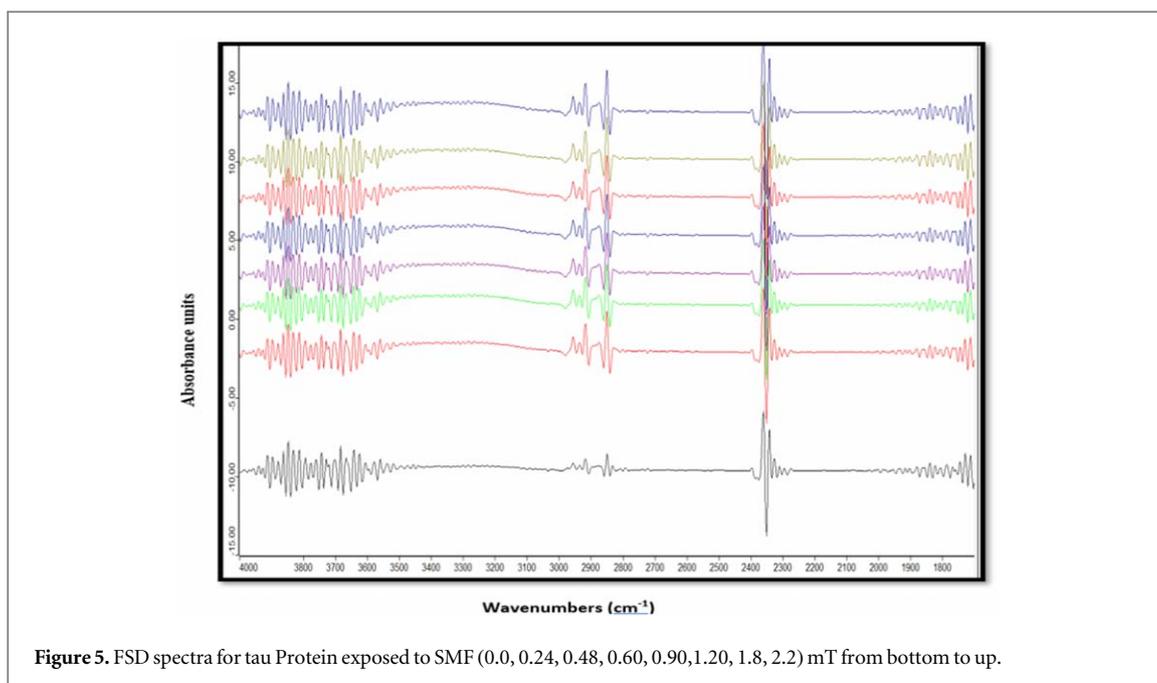
Figure 5. FSD spectra for tau Protein exposed to SMF (0.0, 0.24, 0.48, 0.60, 0.90, 1.20, 1.8, 2.2) mT from bottom to up.

$$\nu = \frac{1}{2\pi}\sqrt{\frac{k}{\mu}} \tag{1}$$

Where ν is the frequency in (cm^{-1}), k is the force constant in N cm^{-1}, and μ is the reduced mass in kg. The only changing variables in the equation are k and μ. These two molecular variables determine the absorbed infrared frequency by any molecule. No two different molecules have the same force constants and atomic masses, so each chemical structure's infrared spectra must differ. It seems that any protein conformation can alter the frequency of the absorbed energy due to modifications in the original bond vibrations of the absorption band. Therefore, it is reasonable to expect that protein aggregation instigates a decrease in the frequency of the secondary structure vibrations [9].

The intensity of the absorption bands in the infrared spectra depends on the concentration of molecules in the sample, as shown by Beer's law in equation (2), which relates the concentration to the absorbance.

$$A = \varepsilon l c \tag{2}$$

Where A is absorbance, ε is absorptivity, ℓ is path length, and c is concentration. Any variation in the number of bonds is directly connected to conformational changes in the protein's structure and can affect absorbance.

Furthermore, the intensities of IR bands can be enhanced by increasing the number of dipole moments of the macromolecules because of their alignment to the applied field. The increase in intensities is proportional to the square of the variation of the dipole moment of the molecule induced by vibrations, as shown by equation (3) [56], the intensity I_k between the vibrational state E_k and the ground state E_0 is

$$I_k = \frac{8\pi^3 N}{3hc(4\pi\varepsilon_0)}(E_k - E_0)\sum_{\alpha=x,y,z}(R_{\alpha k})^2 \tag{3}$$

Where N is Avogadro's number, $R_{\alpha,k}$ is the transition dipole moment between the states 0 and k in the α-direction, E_0, and E_k is the first and the $(k + 1)$ th eigenvalues.

The dipole moment depends on the distance separating the charge; for example, C-H stretching is more intense than C-C rock vibration. The frequencies of stretching vibration are higher than their corresponding bending frequencies, and bonds to hydrogen have higher stretching frequencies than other heavier atoms.

The intensity is related to the substance's temperature and the quantum mechanical interaction between the radiation and the absorber. Therefore, determining absolute intensities is a complicated task. Most researchers calculate the integrated intensities by integrating the area under the absorption bands- to indicate relative changes in the intensities. Any changes in the absorption bands of proteins indicate conformational changes in those proteins due to charge or vibrational changes. The most substantial effect occurs when the magnetic field is perpendicular to the plane of vibration. The most notable variations in the absorbance levels were observed for the symmetric and asymmetric stretching modes of phosphodiester groups of cellular nucleic acids [57]. The overlapping between the absorption bands makes it difficult to determine band intensities accurately. A simple approach is to regard the spectra as a characteristic fingerprint of the conformational change. Therefore,

Table 2. Band positions ($\pm 0.5\ \mathrm{cm}^{-1}$) before and after tau exposure to SMF.

| Peak position at 2.2 mT	Peak position at 0.0 mT	Intensity change	Difference spectra peak position
1710	1710	increase	1708
1730	1730	no change	
1740	1740	no change	1736
1755	1755	increase	1757
1766	1766	no change	
2274	2274	increase	2270
2295	2295	increase	2292
2309	2309	increase	2307
2328	2328	increase	2322
2340	2340	increase	2338
2358	2358	increase	2356
2822	2829	increase	2819
2850	2850	increase	2850
2872	2872	increase	2873
2917	2917	increase	2918
2934	2934	increase	2935
2960	2960	increase	2952
3517	3517	no change	
3536	3536	no change	
3558	3558	no change	
3579	3579	no change	
3607	3607	no change	
3624	3624	no change	
3643	3643	no change	
3667	3667	no change	
3680	3680	no change	
3704	3704	no change	
3714	3714	no change	
3727	3727	no change	
3742	3742	no change	
3759	3759	no change	
3776	3776	no change	
3794	3794	no change	
3813	3813	no change	
3820	3820	no change	
3850	3850	no change	
3860	3860	no change	
3877	3877	no change	
3896	3896	no change	

conformational change in the spectrum may be used to recognize and define transient conformational states of a protein [46].

The absorption spectra revealed several noticeable changes in the amides (IV-VI) regions due to exposure to SMF, as summarized in table 1 and shown in the relevant figures. Most of these bands have shown an increase in the intensity due to exposure to the magnetic field, which implies an increase in the transition dipole moment for these absorption bands. The peak at $512\ \mathrm{cm}^{-1}$ has shown a slight increase in its intensity due to the SMF effect on the NH out-of-plane bending vibrations. The most substantial rise in intensities happened to the bands that involve CO out-of-plane bending with some out-of-plane displacement of the NH group at 595 and $612\ \mathrm{cm}^{-1}$, respectively. A decrease in intensity occurred in the bands $632\ \mathrm{cm}^{-1}$ and $737\ \mathrm{cm}^{-1}$ due to the magnetic field effect on the CO in-plane bending, CC stretch, and CNC deformation [50].

The region $(700\text{–}1200)\ \mathrm{cm}^{-1}$, which composes the fingerprint region, emanates from various vibrations, including out-of-plane N-H bending, C-O, C-C, C-N, and C-H bending vibrations. These bands at 704, 719, 761, 780, 809, 840, 869, 885, 905, 927, 952, 1023, 1043, 1091, 1118, and $1195\ \mathrm{cm}^{-1}$ have increased in intensities due to the SMF exposure, while the bands at 737, 977, 996 and $1147\ \mathrm{cm}^{-1}$ have shown a decrease in their intensities. The increase in intensities happened because of the molecular dipole moment changes, as indicated by equation (3). This change depends mainly on possible variations in the amplitude of vibrations by the induced magnetic force on these molecular vibrations.
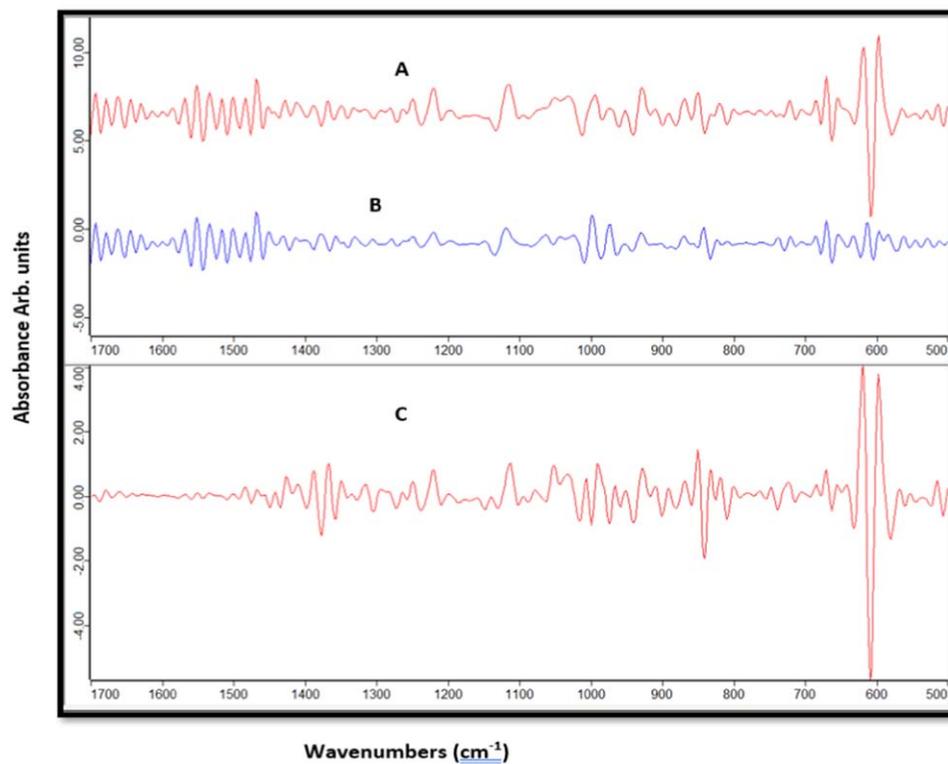
Figure 6. (A) Shows the Spectrum for tau protein exposed to a 2.2mT magnetic field. (B) Shows the spectrum for tau protein at 0.0 mT field. (C) Shows the difference spectrum of (A-B).
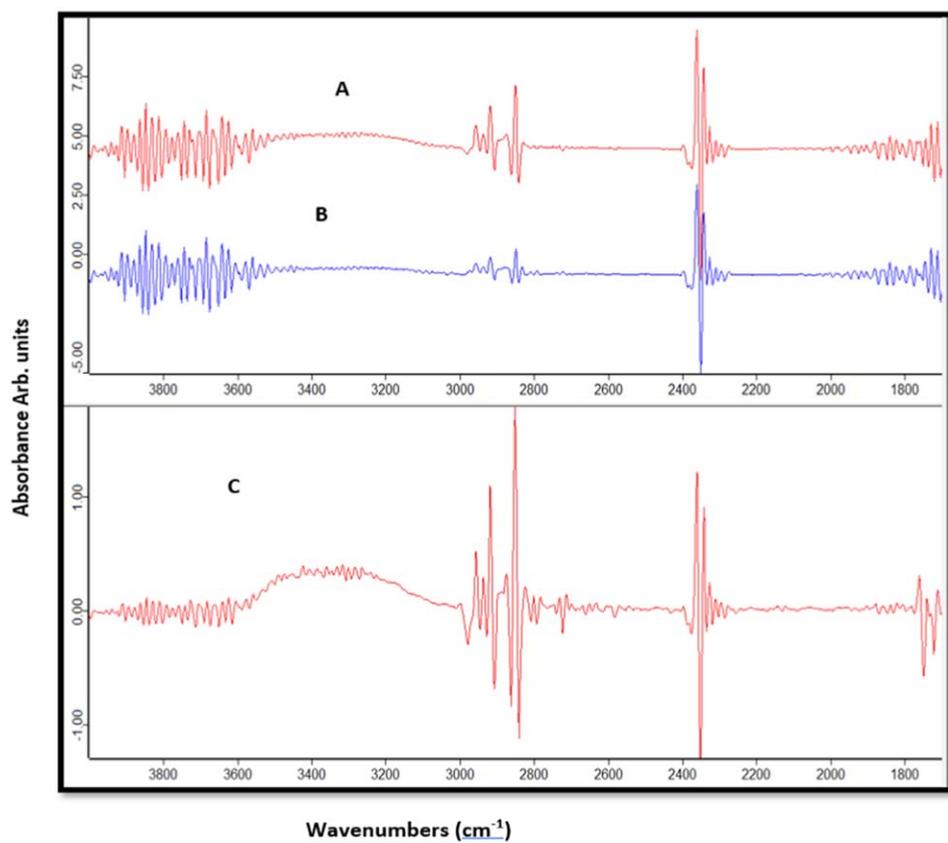
Figure 7. (A) Shows the Spectrum for tau protein after exposure to a 2.2 mT magnetic field. (B) Shows the spectrum for tau protein before any exposure. (C) Is the difference spectrum of (A)–(B).

Furthermore, the subsequent bands showed a shift in their peak positions due to magnetic field exposure in the range $0.0 \rightarrow 2.2$ mT as follows: $(809 \rightarrow 819)$ cm^{-1}, $(823 \rightarrow 832)$ cm^{-1}, $(840 \rightarrow 850)$ cm^{-1}, $(885 \rightarrow 890)$ cm^{-1}, $(905 \rightarrow 910)$ cm^{-1}, $(1023 \rightarrow 1029)$ cm^{-1}, $(1043 \rightarrow 1050)$ cm^{-1}, $(1064 \rightarrow 1075)$ cm^{-1}, and $(1195 \rightarrow 1198)$ cm^{-1}. These peaks shifted to higher energies, which indicate an increase in molecular vibration due to a stronger strain on the vibrating molecules. Nevertheless, few peaks have shifted to lower energies $(996 \rightarrow 993)$ cm^{-1}, $(1118 \rightarrow 1115)$ cm^{-1}, and $(1147 \rightarrow 1143)$ cm^{-1} might be an indication of a decrease in the molecular vibration due to relief of strain on the vibrating molecules.

Tau protein is classified as a natively unfolded protein, and it is expected to have a very low content of secondary structural elements [58, 59]. Therefore, the absorption bands in the amide III region have shown relatively weaker bands compared to the other bands, as shown in figure 4. The band at 1430 cm^{-1} increased in intensity and shifted to 1427 cm^{-1}, while a new weak band seems to be awakened at 1441 cm^{-1} and the band at 1450 cm^{-1} showed a drop in its intensity after tau exposure to SMF.

The absorption bands in amide II and amide I were not affected by the tau exposure to SMF, where little or no changes were observed regarding both intensities and peak positions. The lack of changes can be connected to the low content of secondary structure elements or the small effect on the involved vibrational bonds. It is worth noting that the increase and decrease of intensities should correspond respectively to the peaks and the inverted peaks in the difference spectra in figure 6 and figure 7.

The absorption bands for the range (1700–4000) cm^{-1} are listed in table 2, where they have maintained their positions with little or no intensity increase after exposing tau protein to SMF. Only these bands 2340, 2358, 2829, 2850, 2872, 2917, 2934, and 2960 cm^{-1} have shown a noticeable significant increase in their intensities, out of which the bands at (2872 and 2960) cm^{-1} involve a symmetric and asymmetric stretch of the C-H methyl group, respectively. Also, the bands at (2917 and 2934) cm^{-1} involve the asymmetric stretch of the C-H methylene group, while the band at 2850 cm^{-1} involves the symmetric stretch of the C-H methylene group. These peaks are associated with the presence of saturated lipids, and any intensity increase, mainly CH$_2$ vibrations, can indicate higher content of saturated lipids. An increase in saturated lipids is related to vascular perturbations, which may lead to developing dementia [60–62].

The absorption bands at (2340 and 2358) cm^{-1} have shown a moderate increase in their intensities while maintaining their positions. This is also attributed to changes imposed by the magnetic field on molecular vibrations involving C-C or C-N triple bonds.

The increase in the intensity of the absorption bands because of tau protein exposure to SMF should be proportional to the square of the dipole moment magnitudes of those bands. The transition dipole moment matrix ($R_{\alpha,k}$ in equation (3)) can increase in magnitude by increasing the charge of the dipole moment or the separating distance between the charges; otherwise, all other variables are constant. The exerted magnetic force can induce the above changes in the vibrating atoms of the active molecules.
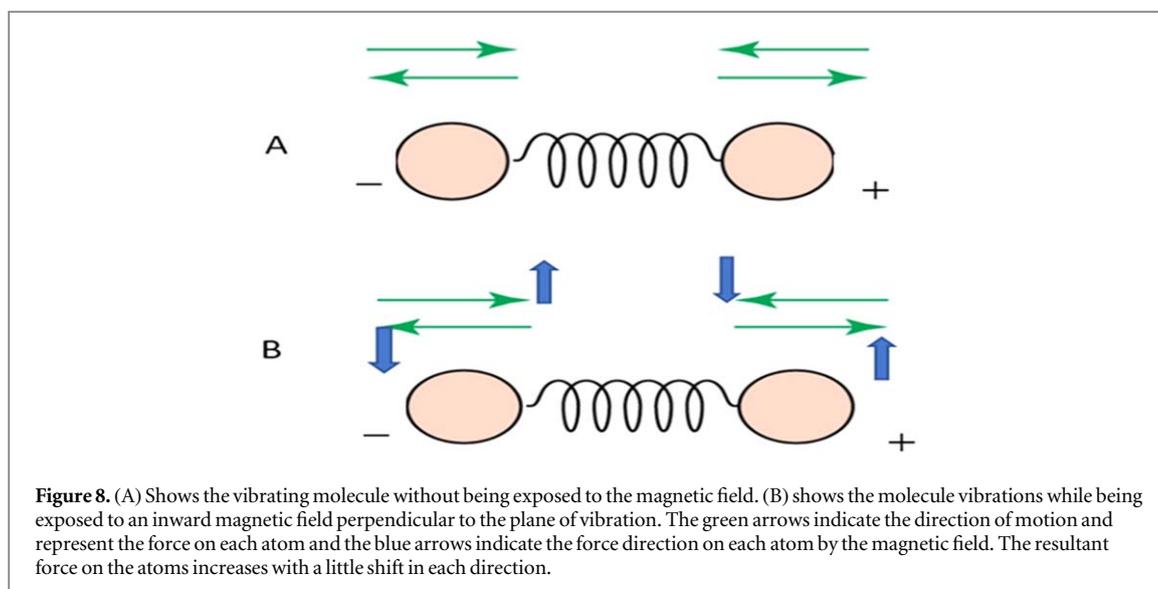
$$F_m = qv \sin \theta \qquad (4)$$

Where q and v are the charge and velocity of the atoms of the active molecule, and θ is the angle between the force and velocity. The magnetic force can exert torque on molecular vibration to align the molecules in the direction of the magnetic field [63]. Moreover, the magnetic force may lead to changes in the amplitude of vibrations and therefore increase or decrease the intensity of the absorption band depending on the molecular orientations with respect to the magnetic field direction. The magnetic field imposes changes on the molecular structure of the protein in the form of aligning and twisting the molecular vibrations, therefore, affecting the bands' intensities. The magnetic force tends to affect asymmetric and out-of-plane motions more than symmetric and in-plane vibrations.

The absorbed molecular frequency may shift because of changes imposed by the magnetic force on molecular vibrations. In other words, any changes in the molecular environment may lead to changes in the spring constant or the displacement of the molecular vibrations affecting the absorbed energy of the molecule. Therefore, exposure of tau protein to SMF can cause changes in the absorbed energy of molecular vibrations and allow some bands to become susceptible to shifting their peak positions.

Our experimental results showed that tau protein exposure to SMF strongly influences the vibrations that involve C-O out-of-plane bending vibration with out-of-plane displacements of the NH group. One may speculate that the effect of the magnetic force on the bending vibrating atoms comes in the form of additional torque twisting the vibrating molecules. Therefore, enhancing the potential energy and kinetic energy of the moving atoms increases the frequency of vibration, and causes the shift in peak position and the increase in intensity.

In the case of amide B, the C-H stretching vibrations are the result of Fermi resonance of the amide II band, which means the frequency has been doubled while the stretching distance is half of that for the fundamental vibrational mode. The magnetic force acts on the vibrating atoms inducing a bending vibration to interfere with the stretching vibration. In other words, if a magnetic force is applied to a vibrating molecule, the magnetic force

Figure 8. (A) Shows the vibrating molecule without being exposed to the magnetic field. (B) shows the molecule vibrations while being exposed to an inward magnetic field perpendicular to the plane of vibration. The green arrows indicate the direction of motion and represent the force on each atom and the blue arrows indicate the force direction on each atom by the magnetic field. The resultant force on the atoms increases with a little shift in each direction.

tends to align the molecules' vibration in its direction. The resulting torque induces an alternating bending effect on the vibrating molecules, which produces changes in the potential energy and the kinetic energy of the vibrating bonds. The magnetic force on moving charges in a magnetic field is expressed by equation (4) and is depicted in figure 8, which shows the induced magnetic forces on each moving charged atom.

The induced magnetic forces impose an out-of-plane bending on the stretched vibration. The resultant force has increased slightly during the stretching out and compressing in with a slight bend in the vibration direction. The increased forces in both directions of the vibrating molecule increase in the amplitude of vibration and yield an intensity increase.

The magnetic force is expected to be more effective against the lighter vibrating molecules such as CH_2 and CH_3, which can be verified from equation (1), where the frequency ν^2 is proportional to $K \mu^{-1}$. Meaning the ν^2 is proportional to the inverse of the reduced mass of the vibrating molecule. Besides, the induced magnetic force has a more proportional impact on vibrations with smaller vibrational amplitudes.

The experimental results have shown that tau protein is sensitive to SMF, reflected by intensity changes and shifts in peak positions at varying degrees for most absorption bands as listed in tables 1 and 2. Tau protein exposure to SMF resembles invoking a molecular environmental change leading to protein conformations through the vibrational changes and other magnetic interactions such as the surface tension generated by the magnetic pressure [64]. It seems that SMF induces molecular polarization in the exposed protein by aligning all molecular vibrations in the plane of the magnetic field direction. The magnetic force interferes by folding all normal protein functions into a two-dimensional surface. These momentary changes in the secondary and tertiary structures of the tau protein leave the possibility open for altering the shape of this sensitive microtubule protein and turning it into an insoluble misfolded protein. On the other hand, it is also possible to use SMF as a noninvasive technique to induce the needed changes to eliminate unwanted protein accumulations.

The intensities' decrease for some bands after the exposure to the magnetic field is caused by a suppressing effect on the amplitude of oscillation which modifies the molecular vibrations in these absorption bands. The inverted peaks in the difference spectra in figures 6 and 7 should also coincide with these bands' positions if there are no shifts. The same is true concerning the increase of intensities where the difference spectra peaks should correspond to the final frequency of the involved bands.

It seems once tau protein is exposed to SMF, a genuine interaction is induced, yielding changes in the protein's secondary and tertiary structures momentarily. The shifts in peak positions correlate to variations in the internal energy of the molecular structure of the protein, which may lead to instantaneous changes in the protein's structure. A magnetic field can affect molecular vibration and induce molecular distortion within the exposed protein. In other words, the magnetic field causes changes to the molecular environment; these induced changes can be helpful in some cases and harmful in others. For example, exposure to a certain strength of magnetic field leading to the unfolding of tau protein into a proper orientation could delay the progression of degenerative diseases. In contrast, exposure to a different type or strength of magnetic field could expedite the aggregation of tau protein and could facilitate the progression of such diseases. Besides, protein exposure to different strengths of magnetic fields and a wide range of electromagnetic radiation may reveal some clues behind protein misfolding or provide a biomarker measurement indicating some disease progression.

The importance of understanding the molecular mechanisms behind protein conformation stems from the need to understand these amyloid aggregates, known for their high heterogeneity and the increasing cases of neurotoxicity. Thus, brain pathogenesis leads to oligomer formations, which is the leading cause of most degenerative neurological diseases, where early detection using invasive techniques could be a lifesaver [37, 65–68]. FTIR spectroscopy is a simple technique to acquire spectra for comparison with a biomarker spectrum of the different neurological diseases to identify each disease. Brain exposure to TSMF can be used as a therapeutic procedure to alter protein conformations and dissolve amyloid oligomers [69]. However, such techniques require a complete understanding of the effects of magnetic fields on protein structure. Besides, more investigations are still needed to understand the mechanisms behind protein misfolding, fibril formation, and aggregation, which leads to most neurological diseases. In general, continued progress in FTIR spectroscopy and imaging, combined with other spectroscopic methods, can lead to an improved mastery, identification, and treatment of different protein-folding diseases [11].

5. Conclusion

The experimental results can be summarized: (1) Tau protein is susceptible to SMF exposure. A significant number of the absorption bands have shown changes in their intensities and peak positions. (2) The magnetic force can affect the molecular vibrations and twist these vibrations, affecting the tau protein's microtubule structure. (3) The shifts in peak positions correspond to changes in the potential energy of the molecular vibrations; these changes are caused by the magnetic field forces on the vibrating atoms. A shift to higher potential energy implies an increase in the stretching amplitude or an increase in the protein force constant leading to a higher frequency in a more solidifying structure. (4) the variations of band intensities relate to the changes in the transition dipole moment or the rate of change for charge in the molecular dipole moment. (5) FTIR spectroscopy has the potential to be used as a sensor for early detections and classifications of dementia diseases through spectrum analysis of a single drop of plasmas. (6) SMF has the potential to be used as a spontaneous therapeutic procedure to impose changes on the molecular environment of the proteins.

Acknowledgments

Data availability statement

The data that support the findings of this study are available upon reasonable request from the authors.

Ethical statement

Not applicable.

Conflict of interest statement

The authors declare no conflict of interest.

ORCID iDs

Saqer M Darwish ⓘ https://orcid.org/0000-0003-0365-5136

References

[1] Kabbara A, El Falou W, Khalil M, Wendling F and Hassan M 2017 The dynamic functional core network of the human brain at rest *Scientific Reports. Nature Publishing Group.* **7** 1 2936
[2] Lozano-Soto E, Soto-León V, Sabbarese S, Ruiz-Alvarez L, Sanchez-Del-Rio M, Aguilar J, Strange B A, Foffani G and Oliviero A 2018 Transcranial static magnetic field stimulation (tSMS) of the visual cortex decreases experimental photophobia *Cephalalgia.* **38** 1493–7
[3] Davila-Péreza P, Pascual-Leonea A and Cudeiro J 2019 Effects of transcranial static magnetic stimulation on motor cortex evaluated by different TMS waveforms and current directions *Neuroscience.* **413** 22–30
[4] Rosen A D 2003 Mechanism of action of moderate-intensity static magnetic fields on biological systems *Cell Biochem Biophys.* **39** 163–73

[5] Lacroix A *et al* 2019 Static magnetic stimulation of the primary motor cortex impairs online but not offline motor sequence learning *Sci Rep.* **9** 1 9886

[6] Dileone M, Mordillo-Mateos L, Oliviero A and Foffani G 2018 Long-lasting effects of transcranial static magnetic field stimulation on motor cortex excitability *Brain Stimulation.* **11** 676–88

[7] Hollis A, Zewdie E, Nettel-Aguirre A, Hilderley A, Kuo H C, Carlson H L and Kirton A 2020 Transcranial static magnetic field stimulation of the motor cortex in children *Frontiers in Neuroscience.* **14** 464

[8] Miyakoshi J 2006 The review of cellular effects of a static magnetic field *Sci. Technol. Adv. Mater.* **7** 305–7

[9] Li H, Lantz R and Du D 2019 Vibrational approach to the dynamics and structure of protein amyloids *Molecules.* **24** 1 186

[10] Calabro E and Magazù S 2013 Unfolding and aggregation of myoglobin can be induced by three hours' exposure to mobile phone microwaves: a FTIR spectroscopy study *Spectrosc. Lett.* **46** 583–9

[11] Miller L 2013 FTIR spectroscopic imaging of protein aggregation in living cells *Biochim. Biophys. Acta* **1828** 2339–46

[12] WHO 1084 Extremely low frequency (ELF) fields, *Environmental Health Criteria 35* (Geneva. Switzerland: World Health Organization)

[13] Falone S, Santini S Jr, Cordone V, Di Emidio G, Tatone C, Cacchio M and Amicarelli F 2018 Extremely low-frequency magnetic fields and redox-responsive pathways linked to cancer drug resistance: insights from co-exposure-based *in vitro* studies *Front Public Health.* **6** 33

[14] Moon J H 2020 Health effects of electromagnetic fields on children *Clin Exp Pediatr.* **63** 422–8

[15] Seomun G, Lee J and Park J 2021 Exposure to extremely low-frequency magnetic fields and childhood cancer: a systematic review and meta-analysis *PLoS One* **14 16** 5 e0251628

[16] Javani Jouni F, Abdolmaleki P and Movahedin M 2013 Investigation on the effect of static magnetic field up to 15 mT on the viability and proliferation rate of rat bone marrow stem cells *In Vitro Cell Dev Biol Anim.* **49** 212–9

[17] Wang Z, Che P L, Du J, Ha B and Yarema K J 2010 Static magnetic field exposure reproduces cellular effects of the Parkinson's disease drug candidate ZM241385 *PLoS One* **5** 11 e13883

[18] Mousavidoust S, Mobasheri H and Riazi G H 2017 Effects of static magnetic fields on the structure, polymerization, and bioelectric of tubulin assemblies *J. Biomol. Struct. Dyn.* **35** 3370–83

[19] Hernando A, Fernando G, García M A, Soto-León V, Alonso-Bonilla C, Aguilar J and Oliviero A 2020 Effects of moderate static magnetic field on neural systems is a noninvasive mechanical stimulation of the brain possible theoretically *Frontiers in Neuroscience.* **14** 419

[20] Calabrò E, Condello S, Magazù S and Lentile R 2011 Static and 50 Hz electromagnetic fields effects on human neuronal-like cells vibration bands in the mid-infrared region *J. Electromagnetic Analysis & Applications.* **3** 69–78

[21] Avila J *et al* 2016 Tau structures *Frontiers in aging neuroscience.* **8** 262

[22] Basu S and Lamprecht R 2018 The role of actin cytoskeleton in dendritic spines in the maintenance of long-term memory *Front Mol Neurosci.* **111** 43

[23] Dadras A, Atarod D, Afrasiabi A, Liaghi A, Naghshineh A and Riazi G 2015 Neuronal spines can be affected by static magnetic fields: the impact on microtubule dynamic nature *JAMSAT.* **1** 86–92

[24] Mietelska-Porowska A, Wasik U, Goras M, Filipek A and Niewiadomska G 2014 Tau protein modifications and interactions: their role in function and dysfunction *Int. J. Mol. Sci.* **15** 4671–713

[25] Chen S, Ge X, Chen Y, Lv N, Liu Z and Yuan W 2013 Advances with RNA interference in Alzheimer's disease research *Drug Des Devel Ther.* **7** 117–25

[26] Lee J C, Kim S J, Hong S and Kim Y 2019 Diagnosis of alzheimer's disease utilizing amyloid and tau as fluid biomarkers *Exp Mol Med.* **51** 1–10

[27] DeTure M A and Dickson D W 2019 The neuropathological diagnosis of alzheimer's disease *Mol Neurodegener.* **14** 1 32

[28] Moloney C M, Lowe V J and Murray M E 2021 Visualization of neurofibrillary tangle maturity in Alzheimer's disease: A clinicopathologic perspective for biomarker research *Alzheimers Dement.* **9** 1554–74

[29] O'Brien R J and Wong P C 2011 Amyloid precursor protein processing and alzheimer's disease *Annu Rev Neurosci.* **341** 85–204

[30] Goedert M and Spillantini M G 2006 A century of alzheimer's disease *Science* **314** 777–81

[31] Despres C *et al* 2017 Identification of the tau phosphorylation pattern that drives its aggregation *Proc Natl Acad Sci U S A.* **114** 9080–5

[32] Serrano-Pozo A, Frosch M P, Masliah E and Hyman B T 2011 Neuropathological alterations in alzheimer disease *Cold Spring Harb Perspect Med.* **1** 1

[33] Pickhardt M, Gazova Z, von Bergen M, Khlistunova I, Wang Y, Hascher A, Mandelkow E M, Biernat J and Mandelkow E 2005 Anthraquinones inhibit tau aggregation and dissolve Alzheimer's paired helical filaments *in vitro* and in cells *J. Biol. Chem.* **280** 3628–35

[34] Schwarz A J *et al* 2018 Alzheimer's disease neuroimaging initiative. topographic staging of tau positron emission tomography images *Alzheimers Dement (Amst).* **10** 221–31

[35] Bejanin A *et al* 2017 Tau pathology and neurodegeneration contribute to cognitive impairment in Alzheimer's disease *Brain.* **140** 3286–300

[36] Liu J and Li L 2019 Targeting autophagy for the treatment of alzheimer's Disease: challenges and opportunities *Front Mol Neurosci.* **12** 203

[37] Braak H and Braak E 1991 Neuropathological stageing of alzheimer-related changes *Acta Neuropathol.* **82** 239–59

[38] Darwish S, Darwish A and Darwish D 2020 An extremely low-frequency magnetic field can affect CREB protein conformation which may have a role in neuronal activities including memory *J. Phys. Commun.* **4** 015009

[39] Darwish S, Alsamamra H, Abushark S, Khalid I, Alfaqeh R and Abuteir M 2017 Effects of extremely low-frequency magnetic field on the secondary structures of β-amyloid and human serum albumin *European Journal of Biophysics.* **5** 89–103

[40] Lai H and Singh N P 1997 Acute exposure to a 60 Hz magnetic field increases DNA strand breaks in rat brain cells *Bioelectromagnetics* **18** 156–65

[41] Elliott A and Ambrose E J 1950 Structure of synthetic polypeptides *Nature* **165** 921–2

[42] Krimm S and Bandekar J 1986 Vibrational spectroscopy and conformation of peptides, polypeptides, and proteins *Adv. Protein Chem.* **38** 181–364

[43] Byler D M and Susi H 1986 Examination of the secondary structure of proteins by deconvolved FTIR spectra, *Biopolymer.* **25** 469–87

[44] Derkus B, Ozkan M, Emregul K C and Emregul E 2016 Single frequency analysis for clinical immunosensor design *RSCAdv.* **6** 281

[45] Goedert M, Spillantini M G, Jakes R, Rutherford D and Crowther R A 1989 Multiple isoforms of human microtubule-associated protein tau: sequences and localization in neurofibrillary tangles of alzheimer's disease *Neuron* **3** 519–26

[46] Barth A 2007 Review of infrared spectroscopy of protein *Biochim. Biophys. Acta* **1767** 1073–101

[47] Kong J and Yu S 2007 Fourier transform infrared spectroscopic analysis of protein secondary structures *Acta Biochim. Biophy. Sin.* **39** 549–59

[48] Singh B R, DeOliveira D B, Fu F and Fuller M P 1993 Fourier transform infrared analysis of amide III bands of proteins for the secondary structure estimation *Proc. SPIE 1890, Biomolecular Spectroscopy III.* 49, 55

[49] Garidel P and Schott H 2006 Fourier-transform midinfrared spectroscopy for analysis and screening of liquid protein formulations Part 2 : detailed analysis and applications *Bioprocess Int.* **40** 48–55

[50] Yatsyna V *et al* 2016 'Far-infrared amide IV-VI spectroscopy of isolated 2- and 4-Methylacetanilide' *J. Chem. Phys.* **145** 104309

[51] Kozłowicz K, Różyło R, Gładyszewska B, Matwijczuk A, Gładyszewski G, Chocyk D, Samborska K, Piekut J and Smolewska M 2020 Identification of sugars and phenolic compounds in honey powders with the use of GC-MS, FTIR spectroscopy, and x-ray diffraction *Sci Rep.* **10** 1 16269

[52] Deleris G and Petibios C 2003 Applications of FT-IR spectrometry to plasma contents analysis and monitoring *Vib Spectroscopy.* **32** 129–36

[53] Brain C S 2019 Organic nitrogen compounds IV: nitriles *Spectroscopy.* **34** 18–21

[54] Zhuang J, Li M, Pu Y, Ragauskas A J and Yoo C G 2020 Observation of potential contaminants in processed biomass using Fourier transform infrared spectroscopy *Applied Sciences.* **10** 12 4345

[55] Güler G, Gärtner R M, Ziegler C and Mäntele W 2016 Lipid-protein interactions in the regulated betaine symporter BetP probed by infrared spectroscopy *J. Biol. Chem.* **291** 4295–307

[56] Burcl R, Carter S and Handy N C 2003 Infrared intensities from the multimode code *Chem. Phys. Lett.* **380** 237–44

[57] Simonova D and Karamancheva I 2013 Application of Fourier transform infrared spectroscopy for tumor diagnosis *BBE.* **27** 4200–7

[58] Schweers O, Schonbrunn-Hanebeck E, Man A and Mandelko E 1994 Structural studies of tau protein and alzheimer paired helical filaments show no evidence for p-structure *J. Biol. Chem.* **269** 24290–7

[59] Jeganathan S, von Bergen M, Mandelkow E M and Mandelkow E 2008 The natively unfolded character of tau and its aggregation to Alzheimer-like paired helical filaments *Biochemistry.* **47** 10526–39

[60] Correia M *et al* 2016 FTIR spectroscopy - a potential tool to identify metabolic changes in dementia patients *J Alzheimer's Neurodegener Dis.* **2** 007

[61] Tamm L K and Tatulian S A 1997 Infrared spectroscopy of proteins and peptides in lipid bilayers *Q. Rev. Biophys.* **30** 365–429

[62] Dritsa V 2012 FTIR spectroscopy in medicine. infrared spectroscopy-life and biomedical sciences *Intech Open, Rijeka, Croatia.* **16** 271–88

[63] Calabrò E and Magazù S 2017 The α-helix alignment of proteins in water solution toward a high-frequency electromagnetic field: a FTIR spectroscopy study *Electromagnetic Biology and Medicine.* **36** 279–88

[64] Ghodbane S, Lahbib A, Sakly M and Abdelmelek H 2013 Bioeffects of static magnetic fields: oxidative stress, genotoxic effects, and cancer studies *BioMed Res. Int.* **2013** 602987

[65] Devitt G *et al* 2021 Conformational fingerprinting of tau variants and strains by Raman spectroscopy *RSC Adv.* **11** 8899–915

[66] Lois C, Gonzalez I, Johnson K A and Price J C 2019 PET imaging of tau protein targets: a methodology perspective *Brain Imaging Behav.* **13** 333–44

[67] Carmona P, Molina M, Calero M, Bermejo-Pareja F, Martínez-Martín P and Toledano A 2013 Discrimination analysis of blood plasma associated with alzheimer's disease using vibrational spectroscopy *J. Alzheimers Dis.* **34** 911–20

[68] Zetterberg H and Blennow K 2021 Moving fluid biomarkers for alzheimer's disease from research tools to routine clinical diagnostics *Mol Neurodegeneration.* **16** 10

[69] Perez F P, Maloney B, Chopra N, Morisaki J J and Lahiri D K 2021 Repeated electromagnetic field stimulation lowers amyloid-β peptide levels in primary human mixed brain tissue cultures *Sci Rep.* **11** 1 621