

EFFECT OF 50 HZ MAGNETIC FIELD ON THE STRUCTURE OF BOVINE SERUM ALBUMIN

I.M. KHATER

Biophysics Department, Faculty of Science, Cairo University, Giza, 12613, Egypt,
ibrahim.khater@hotmail.com

Abstract. Bovine serum albumin (BSA), in the solid state, was exposed to 50 Hz magnetic field of 10 gauss (1 mT) and 20 gauss (2 mT) for one hour. The changes in the secondary and tertiary structure of BSA were investigated. The Fourier transform infrared spectroscopy (FTIR) indicated that there is a relative decrease in absorption intensity of the amide I and amide II bands for the BSA exposed to 20 gauss 50 Hz magnetic field. The second derivative and multi-peak fitting of the amide I peak of the resultant spectrum indicated that there is a relative variation in the secondary structure of BSA exposed to 20 gauss magnetic field.

Key words: electromagnetic radiation, health hazards, proteins, FTIR, secondary structure, 3D structure.

INTRODUCTION

In recent years it has been discovered that exposure to low frequency magnetic fields may lead to changes in activity of biological systems both *in vivo* and *in vitro*. The present work is studying the effect of alternating magnetic field on a structural state of BSA.

Bovine serum albumin (BSA) is the most abundant protein in plasma contributing to osmotic blood pressure. It plays important roles in the transport, distribution and metabolism of many exogenous ligands, including fatty acids, amino acids, drugs and pharmaceuticals [5, 7, 9, 12].

Bovine serum albumin (BSA) is a single-chain 582 amino acid globular nonglycoprotein cross-linked with 17 cystine residues (8 disulfide bonds and 1 free thiol). BSA is divided into three linearly arranged, structurally distinct, and evolutionarily related domains (I–III) [1, 3, 13, 14]; each domain is composed of two sub-domains (A, B). BSA has two tryptophans embedded in two different

Received: January 2013;
in final form February 2013.

domains, one is Trp-134, located in the proximity of the protein surface, but buried in a hydrophobic pocket of domain I, and the other is Trp-214, located in an internal part of domain (II).

MATERIALS AND METHODS

BSA was purchased from Sigma Chemical (St. Louis, MO, USA) and used without further purification. Protein concentration was determined by measuring the optical density at 280 nm.

BSA was irradiated at room temperature, using an electrical magnet, manufactured at Cairo University, to 10 gauss and 20 gauss 50 Hz magnetic field for one hour.

Films with a small amount of BSA sample were used to measure the FTIR spectra. The spectra were recorded from 2000 to 1000 cm^{-1} on an FTIR spectrometer (Jasco V570; Easton, MD, USA). The second-derivative spectra and the multi-peak fitting of the amide I bands are obtained using the Origin software Version 6.0. The difference spectra are obtained by subtracting the second-derivative spectra of different samples from control.

RESULTS AND DISCUSSION

FTIR is a reliable technique for the determination of some main features of the secondary structure of protein in solution. The analysis of the amide I band (1700–1600 cm^{-1}) originating from C=O stretching vibration of the peptide group would yield useful information about the conformation of protein [9, 11, 15]. In the present work, FTIR is employed in order to monitor the secondary structure of native and exposed BSA.

Fig. 1 presents the area normalized FTIR spectra of native (control) BSA and exposed samples (from 1000 to 2000 cm^{-1}). The amide I and amide II peaks show an increase in the absorption of both peaks for 10 gauss and 20 gauss exposed samples compared to control.

Fig. 2 presents the second-derivative spectra of the amide I region (from 1600 to 1700 cm^{-1}). The second-derivative spectra are a band narrowing technique which would help to enhance the resolution of the overlapping bands contributing to the amide I band. Fig. 3 presents the difference spectra (second-derivative of exposed subtracted from control).

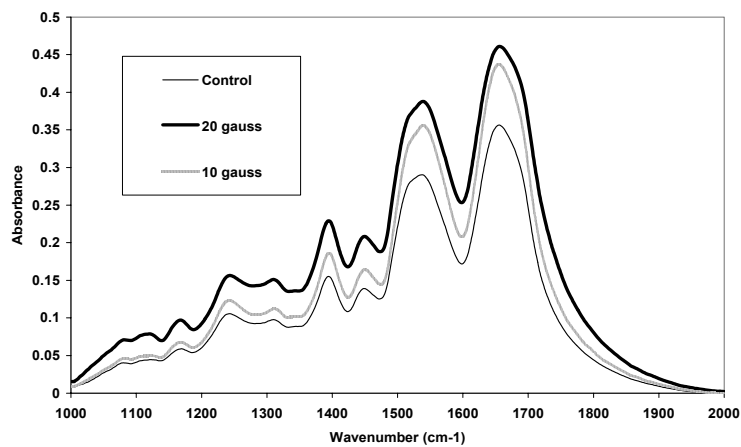


Fig.1. FTIR spectra of native (control) BSA and exposed samples from 1000 to 2000 cm^{-1} .

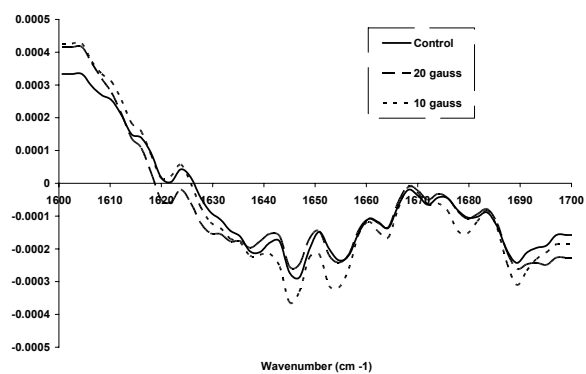


Fig. 2. The second-derivative spectra of the amide I region of FTIR spectra of native (control) BSA and exposed samples from 1600 to 1700 cm^{-1} .

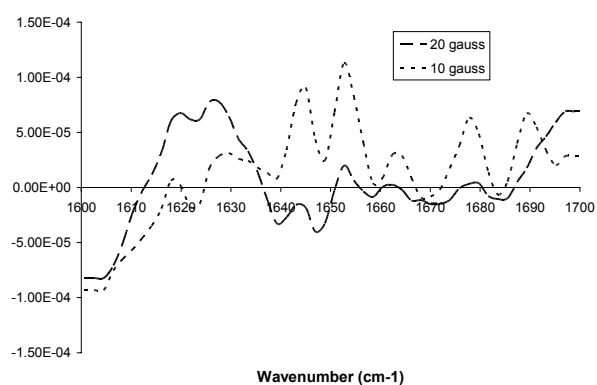


Fig. 3. The difference spectra (second-derivative of exposed subtracted from control).

Tables 1, 2 and 3 present the multi-peaks curve fitting of the amide I region (from 1600 to 1700 cm^{-1}) by Origin software Version 6.0 for native BSA, 10 gauss exposed BSA and 20 gauss exposed BSA. The positions of peaks, peaks areas and area percentage for each peak are included. Tables show that a few changes in the content of BSA secondary structure induced by the 10 gauss magnetic field while the content of BSA secondary structure before and after being exposed to 20 gauss magnetic field varied significantly.

Table 1

Presents the multi-peaks curve fitting of the amide I region (from 1600 to 1700 cm^{-1}) for control BSA

Peak	Center (cm^{-1})	Area (cm^2)	%
1 turns	1693.9	0.0224	16.917
2 turns	1676.3	0.0217	16.407
3 Turns	1664.7	0.0223	16.864
4 Helix	1653.3	0.0225	17.018
5 Intra-molecular β sheet	1640.5	0.0272	20.532
6 Intermolecular β sheet	1625.8	0.0162	12.257

Table 2

Presents the multi-peaks curve fitting of the amide I region (from 1600 to 1700 cm^{-1}) for 10 gauss exposed BSA

Peak	Center (cm^{-1})	Area (cm^2)	%
1 turns	1693.6	0.0224	16.632
2 turns	1676.5	0.0218	16.140
3 Turns	1667.9	0.022	17.001
4 Helix	1651.4	0.0230	17.039
5 Intra-molecular β sheet	1641.7	0.0281	20.853
6 Intermolecular β sheet	1626.2	0.0166	12.331

Table 3

Presents the multi-peaks curve fitting of the amide I region (from 1600 to 1700 cm^{-1}) for 20 gauss exposed BSA

Peak	Center (cm^{-1})	Area (cm^2)	%
1 turns	1693.1	0.0174	16.5633
2 turns	1686.2	0.0157	14.8803
3 Turns	1669.4	0.0191	18.1241
4 Helix	1652.9	0.0189	17.9403
5 Intra-molecular β sheet	1640.9	0.0229	21.7338
6 Intermolecular β sheet	1624.7	0.0113	10.7579

The contents of intermolecular β -sheets decreased at 1625 cm^{-1} after the exposure to 20 gauss magnetic field and its value fell from 12.25% to 10.75%. Oppositely the contents of intra-molecular β -sheets at 1640 cm^{-1} increased from 20.5% to 21.7%. The total percentage of β -sheets almost unchanged suggested that a structural transition from intermolecular β -sheets to intra-molecular ones occurred [5]. The content of α -helices at 1653 cm^{-1} increased slightly from 17% to 17.9% [2]. The content of turns at 1664 cm^{-1} increased from 17% to 18.1% while the turns at 1667 cm^{-1} decreased from 16.4% to 14.88% but that at 1693 remained approximately unchanged.

CONCLUSION

Although the primary mechanisms of action of a magnetic field on biological systems are unknown [3], the spectral data presented in this paper confirm the changes occurring in a magnetic field in the system of hydrogen bonds [16]. Magnetic field may increase the number of hydrogen bonds [4] and may also rearrange them which may lead to structural changes of proteins.

REFERENCES

1. BROWN, K.F, M.J. CROOKS, Displacement of tolbutamide, glibenclamide and chlorpropamide from serum albumin by anionic drugs, *Biochemical Pharmacology*, 1979, **25**, 1175–1178.
2. BYLER, D.M., J.N. BROUILLETTE, H. SUSI, Quantitative studies of protein structure by FTIR spectral deconvolution and curve fitting, *Spectroscopy*, 1986, **3**, 29–32.
3. CARTER, D.C, J.X. Ho, Structure of serum albumin, *Advances in Protein Chemistry*, 1994, **45**, 153–203.
4. CHANG, K., C. WENG, The effect of an external magnetic field on the structure of liquid water using molecular dynamics stimulation, *Journal Applied Physics*, 2006, **100**, 043917-1–043917-6.
5. DONG, A., B. KENDRICK, L. KREILGÅRD, J. MATSUURA, M.C. MANNING, J.F. CARPENTER, Spectroscopic study of secondary structure and thermal denaturation of recombinant human factor XIII in aqueous solution, *Archives of Biochemistry and Biophysics*, 1997, **47**, 213–220.
6. FENG, X.Z., Z. LIN, L.J YANG, C. WANG, C.L. BAI, Investigation of the interaction between acridine orange and bovine serum albumin, *Talanta*, 1998, **47**, 1223–1229.
7. HE, X.M., D.C. CARTER, Atomic structure and chemistry of human serum albumin, *Nature*, 1992, **358**, 209–215.
8. KUMR, C.V., A. BURANAPRAPUK, Tuning the selectivity of protein photocleavage: spectroscopic and photochemical studies, *Journal of the American Chemical Society*, 1999, **121**, 4262–4270
9. MARIO, S., M. MICHELA, M. TANIA, C. LAURA, M. GIULIA, C. RAFFAELLA, D. LUISA, T. FLORIAN, M. LUCA, M. GIOVANNA, J KOURIE JOSEPH M., B. ORSO, S. DEEPAK, I. HIDEYO, A. KIRSCHNER DANIEL, G. FORLONI, T. FABRIZIO, Structural properties of Gerstmann-Straussler-Scheinker disease amyloid protein, *The Journal of Biological Chemistry*, 2003, **278**, 48146–48153.

10. MOON, S., K. B. SONG, Effect of γ -irradiation on the molecular properties of egg white proteins, *Food Science and Biotechnology*, 2000, **9**, 239–242.
11. NATALELLO, A., D. AMI, S. BROCCA, M. LOTTI, S.M. DOGLIA, Secondary structure, conformational stability and glycosylation of a recombinant *Candida rugosa* lipase studied by Fourier-transform infrared spectroscopy, *The Biochemical Journal*, 2005, **385**, 511–517.
12. PANJEHSHAHIN, M.R., C.J. BOWMER, M.S. YATES, A pitfall in the use of double-reciprocal plots to estimate the intrinsic molar fluorescence of ligands bound to albumin, *Biochemical Pharmacology*, 1989, **38**, 155–159.
13. PETERS, T., *All About Albumin: Biochemistry, Genetics and Medical Applications*, Academic Press, San Diego, 1995.
14. ROSENOER, V.M., M. ORATZ, M. A. ROTHSCHILD, *Albumin Structure, Function and Uses*, Pergamon Press, N.Y., 1977.
15. WEERT, M., DE. VAN, I. HARIS PARVEZ, E. HENNINK WIM, J.A. CROMMELIN DAAN, Fourier transform infrared spectrometric analysis of protein conformation: effect of sampling methods and stress factors, *Analytical Biochemistry*, 2001, **297**, 160–169.
16. ZALESSKAYA, G.A., N.P. MIT'KOVSKAYA, O.A. GALAI, A.V. KUCHINSKII, O.V. LASKINA, Change in the absorption spectra of blood exposed to low frequency magnetic field, *Zhurnal Prikladnoi Spektroskopii*, 2007, **74**, 199–204.