Linear and thermo-optically generated nonlinear optical response of bovine serum albumin and its constituent amino acids in continuous wave z-scan

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ABSTRACT

Proteins are large biomolecules in the form of polypeptide chains consisting of amino acid (AA) residues. Ultraviolet–visible absorption spectroscopy and continuous wave (CW) z-scan of bovine serum albumin (BSA) and some of its constituent AAs were examined to deduce the relationship between the optical properties of this protein molecule and its constituents. From the analysis of their optical spectra, the absorption at 278 nm by BSA is found to be the outcome of the cumulative effects of the absorptions by constituent aromatic AA residues, cysteine disulfide bonds, and methionine. Similarly, the closed aperture CW z-scan of BSA and those of the constituent AAs at 74–106 mW incident optical power at 655 nm indicate that thermally generated third-order optical effects arise in BSA and its aromatic AA residues due to multiphoton absorptions. The nonlinear optical (NLO) responses of BSA and those of the AA residues are compared in terms of their molar phase shift per unit power, which indicate a possible relationship between the NLO property of BSA and its AA residues.

I. INTRODUCTION

Amino acids are protein-forming biomolecules and are the building blocks of life. All of the 21 protein-forming residues were discovered from the hydrolysis of proteins within the early 19th century to early 20th century, the first discovered one being glycine in 1820 and the last one being methionine in 1922.[1] The study of amino acids and proteins using spectroscopic techniques dates back to the late 19th century with Soret's ultraviolet–visible (UV–vis) absorption study of blood in 1883.[2] This technique is based on the absorption of electromagnetic (EM) radiation by atoms or molecules. Absorption of photons in the ultraviolet and visible regions of the EM spectrum may result in $\pi \rightarrow \pi^*$, $n \rightarrow \pi^*$, $\pi \rightarrow \sigma^*$, $n \rightarrow \sigma^*$, or even ionization of the molecule. The extent of absorption at a particular wavelength is expressed in terms of the molar extinction coefficient (ε_M) expressed in units of $L\ \mathrm{mol}^{-1}\ \mathrm{cm}^{-1}$ $\left(\mathrm{M}^{-1}\mathrm{cm}^{-1}\right)$. Biomolecules, such as proteins, amino acids, and nucleic acids, absorb light in the near UV (150–400 nm) and visible regions (400–800 nm) of the electromagnetic spectrum.[3] These absorptions in protein molecules are due to their constituent aromatic amino acid (AA) residues; for example, tryptophan absorbs at around 280 nm $\left(5390\ \mathrm{M}^{-1}\mathrm{cm}^{-1}\right)$, tyrosine absorbs at 275 nm $\left(1390\ \mathrm{M}^{-1}\mathrm{cm}^{-1}\right)$, and phenylalanine absorbs at around 257 nm $\left(195\ \mathrm{M}^{-1}\ \mathrm{cm}^{-1}\right)$.[4,5] All of these absorptions correspond to $\pi - \pi^*$ transition and are accompanied by vibrational fine structures appearing as shoulders alongside the peaks.[6] In addition, cysteine disulfide bonds contribute weakly around 260 nm $\left(125\ \mathrm{M}^{-1}\mathrm{cm}^{-1}\right)$.[3] Furthermore, the amide chromophores in peptide links of the protein molecules exhibit absorption around the 190–220 nm region.[7] The position and shape of the absorption bands of a protein molecule reflect its structural and environmental features.[6,8]