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Thermodynamics and binding mechanism of polyphenon-60 with human lysozyme elucidated by calorimetric and spectroscopic techniques

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ABSTRACT

Protein-drug interaction offer information of the structural features that determine the therapeutic effectiveness of drug and have become an attractive research field in life science, chemistry, and clinical medicine. Interaction of pharmacologically important antioxidant drug polyphenon-60 with human lysozyme (Lys) at physiological pH 7.4 has been studied by using calorimetric and various spectroscopic techniques. UV-visible spectroscopy results indicate the complex formation between Lys and polyphenon-60. The binding constant, quenching mechanism and the number of binding sites were determined by the fluorescence quenching spectra of Lys in presence of polyphenon-60. Fluorescence data calculate that the polyphenon-60 interact with Lys through static quenching mechanism with binding affinity of $2.9 \times 10^4 \text{ M}^{-1}$. The average binding distance between drug and Lys was found to be 2.89 nm on the basis of the theory of Förster's energy transfer. Isothermal titration calorimetry (ITC) data reveals the thermodynamic investigations which suggest that the interaction of Lys and polyphenon-60 through exothermic process and enthalpy driven and also explore that the polyphenon-60 binds in both site of Lys with high and low affinity. Hydrogen bonding (high affinity) and hydrophobic interactions (low affinity) are the major forces in stabilizing the drug protein complex. Far-UV CD and FTIR results decipher the conformational alterations in the secondary structure of Lys.

Keywords: Circular dichroism; Fluorescence spectroscopy; Human lysozyme; Isothermal titration calorimetry; Polyphenon-60.

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