ABSTRACT

The principal focus of the thesis is exploration of electrostatic interaction of food dyes with bovine and human serum albumins (BSA and HSA, respectively) and small molecular assemblies. In this regard various azo food dyes are studied. Here in, observation of inversion in Stern-Volmer (S-V) quenching plot of carmoisine by diheptyl viologen has been reported in neat methanol for the first time, on the other hand structurally similar methyl viologen shows saturation. The cause of quenching is attributed to electrostatic binding of the dye with viologens and their aggregates in the ground state. This study has been utilized to probe aggregation of viologens fluorimetrically in neat methanol solvents. There are various methods available in literature for the determination of the critical micellar concentration (CMC) of surfactant, which are based on surface tension measurement, micellar catalysis and spectroscopy. A new approach to follow surfactant aggregation and CMC has been formulated in this thesis employing reorganization energy calculation by using a solvent-sensitive fluorescent probe. A detailed account of electrostatic binding of cochineal red A (CR) with BSA and HSA is presented using optical spectroscopic techniques and molecular docking explorations. Also involvement of two binding sites in BSA has resulted in a pH-dependent texture of S-V quenching plot. Ligand binding pockets (sites) of proteins may have pH-switchable amino acid side chains, e.g., positively charged histidine side chain. The effect of modification of charge of the binding site due to pH alterations on the affinity has been quantified by Brønsted–Bjerrum equation-like expression. It has been shown for the first time that, for electrostatic binding of dyes in subdomain IIA of HSA, by using such expression one can find out the effective charge of the protein pocket and possible role of charge of histidine side-chain on the affinity. The study has revealed that the warfarin binding site (subdomain IIA of HSA) is positively charged. It has been shown with tartrazine (TZ)-BSA and TZ-HSA binding studies that how salts can alter the magnitude of thermodynamic parameters. In this regard ionic strength variations have shown an evolution of sign and magnitude of thermodynamic parameters, which has indicated overshadowing of electrostatic forces in high salt concentrations. On the application side, salt induced removal of dye could be important with respect to protein-dye affinity chromatography.

Keywords: serum albumin, azo food dye, fluorescence quenching, electrostatic interaction, docking, binding affinity, ionic strength, molecular aggregates.

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