Supporting Informations

Spectroscopic Investigation of the Effects of Salt on the Binding of Tartrazine with Two Homologous Serum Albumins: Quantification by Use of Debye-Hückel Limiting Law and Observation of Enthalpy-Entropy Compensation

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Quantum yield of tartrazine

The quantum yield of tartrazine is determined in water taking coumarin-314 as a standard $(\Phi_f = 0.68)$ in ethanol¹ using following equation

$$\Phi = \Phi_R \frac{I}{I_R} \frac{OD_R}{OD} \frac{n^2}{n_R^2}$$

where Φ is the quantum yield, *I* is the integrated intensity, OD is the optical density, and *n* is the refractive index. The subscript *R* refers to the reference fluorophore of known quantum yield. The quantum yield of tartrazine is determined to be $(6.09 \pm 0.06) \times 10^{-5}$ at 236 nm at 298K in water.

Solubility:

The solubility of tartrazine is 120g/lit at 25°C in water.

Normalized fluorescence spectra of BSA ($\lambda_{ex} = 295 \text{ nm}$) and Tartrazine ($\lambda_{ex} = 427 \text{ nm}$) in 5 mM phosphate buffer of pH 7.4 at 298K. [BSA] = 2 μ M and [Tartrazine] = 12 μ M



Fluorescence spectra of SA as a function of Tartrazine concentration in 5 mM phosphate buffer at pH7.4, (A) BSA, (B) HSA. $\lambda_{ex} = 295$ nm at 300K.





Effect of selected site markers on tartrazine-HSA in 5 mM phosphate buffer of pH 7.4 at 298K. [HSA] = 2 μ M and [protein]:[site marker] = 1:1. Symbols with cap include error bar.



Job's plots of tartrazine-HSA and BSA systems in 5 mM phosphate buffer of pH 7.4 in presence of 0.1 M NaCl at 298K. $C^0 = 1.2 \times 10^{-5}$ M.



Table S1

Accessible surface area (ASA) in $Å^2$ of interacting residues of BSA (uncomplexed) and in their complex with Tartrazine (TZ).

Subdomain IIA				Subdomain IIIA			
Residues		BSA ASA(Å ²)	BSA-TZ ASA(Å ²)	Residues		BSA ASA(Å ²)	BSA-TZ ASA(Å ²)
Leu197	Helix	41	7	Glu443	Turn	93	83
Arg198	Helix	63	50	Pro446	Helix	20	2
Ser201	Helix	17	11	Cys447	Helix	41	37
Ala209	Helix	32	30	Asp450	Helix	43	3
Trp213	Helix	76	27	Ser453	Helix	20	4
Arg217	Helix	45	8	Leu454	Helix	8	6
Gln220	Helix	5	5	Leu480	Coil	39	15
Val292	Coil	35	24	Val481	Turn	33	26
				Arg483	Helix	6	3

Table S2

Accessible surface area (ASA) in $Å^2$ of interacting residues of HSA (uncomplexed) and in their complex with Tartrazine (TZ).

Subdomain IIA				Subdomain IIIA			
Residues		HSA	HSA-TZ	Residues		HSA	HSA-TZ
		ASA(Å ²)	ASA(Å ²)			ASA(Å ²)	ASA(Å ²)
Lys199	Helix	37	13	Lys436	Helix	105	84
Trp214	Helix	66	40	His440	Coil	53	39
Arg218	Helix	42	17	Pro447	Helix	26	20
Leu219	Helix	18	14	Cys448	Helix	42	7
Arg222	Helix	37	11	Asp451	Helix	32	8
Leu238	Helix	31	10	Tyr452	Helix	36	32
His242	Helix	5	0				
Arg257	Helix	22	18				
Ala291	Helix	36	4				
Glu292	Turn	95	85				

1. Reynolds, G. A.; Drexhage, K. H. Optics Commun. 1975, 13, 222-225.