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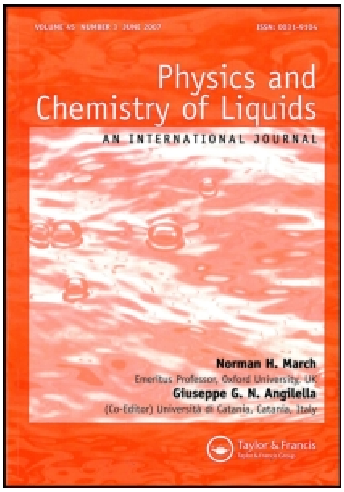

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Spectroscopic analysis of naphtholazobenzimidazole in organised solution

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Micelle–water partition coefficient (K_x) of naphtholazobenzimidazole dye (NAB) in aqueous solutions of cetyltrimethylammonium bromide (CTAB) and sodium dodecyl sulphate (SDS) has been determined spectrophotometerically. Changes in absorption patterns of dye caused by surfactants and solvents were quantified in terms of dye–surfactant ratio (n_D/n_S) and solvent water partition coefficients (P), respectively. Multiple residence sites have been suggested for dye molecules within micelles, based on shifts in azo-hydrazone tautomeric equilibrium. Micelle–water partition coefficients were used to evaluate the influence of dye on critical micelle concentration of CTAB and SDS. At same micelle concentration, M, the relative solubility of NAB was greater in cationic surfactant CTAB than in anionic surfactant SDS.

Keywords: dye–surfactant interaction; naphtholazobenzimidazole; partition coefficient

1. Introduction

Surfactant-dependent optical properties of dye offer an opportunity to study surfactant–dye interactions [1–17]. Surfactant-induced changes in spectral absorption have frequently been used to highlight various factors that contribute towards the growth of surfactant–dye aggregates [18–26]. The binding behaviour of these aggregates provides the key to manufacturing of nanostructured materials [27,28]. The physical behaviour of surfactant micelles can be visualised as a pseudo model to mimic drug–membrane interactions [29]. Lipid–water distribution coefficient is more critical than lipid solubilisation alone for absorption of most drugs [30] and is highly dependent on drug size and polarity [31]. Previously, we reported the effect of dyes hydrophobicity on the distribution of hemicyanine dyes between aqueous and micellar pseudo phase [10,19,32–34]. This work examines the spectroscopic response of naphtholazobenzimidazole (NAB) dye, that is, 2-methyl-5-(diazo-2-naphthol)-benzimidazole as a function of surfactant concentration, ranging from pre-micellar to micellar region in aqueous medium. NAB exists almost entirely in hydrazone form

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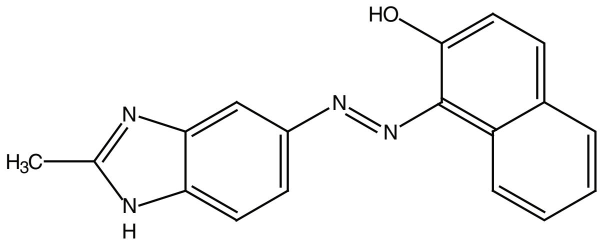
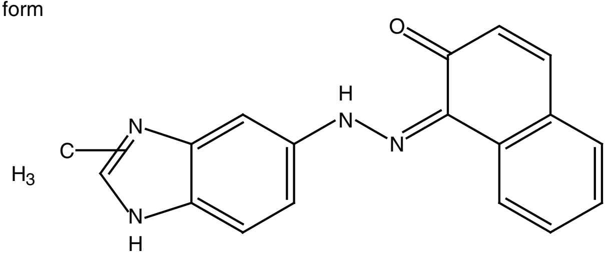


Azo form



Hydrazone form

Figure 1. Naphtholazobenzimidazole dye (NAB).

($\lambda_{max} = 495$ nm) in aqueous solution and tuatomerises to corresponding azo form ($\lambda_{max} = 418$ nm) in organised molecular assemblies of ionic surfactants (Figure 1).

Difference spectra of azo dye obtained in sodium dodecyl sulfate (SDS) and cetyltrimethylammonium bromide (CTAB) solutions were used for evaluating partition equilibrium constants (K_c), and relative solubility (S_t/S_o). Spectral changes occurring in solvents and surfactants were utilised to demonstrate dye–surfactant associations, aggregation tendency and proclivity towards microenvironment.

2. Experimental

2.1. *Materials*

Cetyltrimethylammonium bromide and SDS were the products of Fluka and Sigma, respectively. They were used without further purification. Naphtholazobenzimidazole was prepared by diazo coupling of 5-amino benzimidazole with 2-naphthol, adopting a reported method [35]. Crystallisation from methanol yielded scarlet crystals [λ_{max} (H_2O) = 495 nm]. The characterisation was done by TLC, ^{1}H-NMR and UV-vis absorption measurements. Solvents utilised for partitioning study were obtained from Aldrich and used as received.

2.2. *Measurements*

2.2.1. *Critical micelle concentration*

Measurement of electrical conductivity of CTAB and SDS was made by using a Microprocessor conductivity meter of WTW (model LF 2000/C) at 25 ± 0.01°C. The critical micelle concentration (cmc) was determined from the plot of conductivity *versus* surfactant concentration.

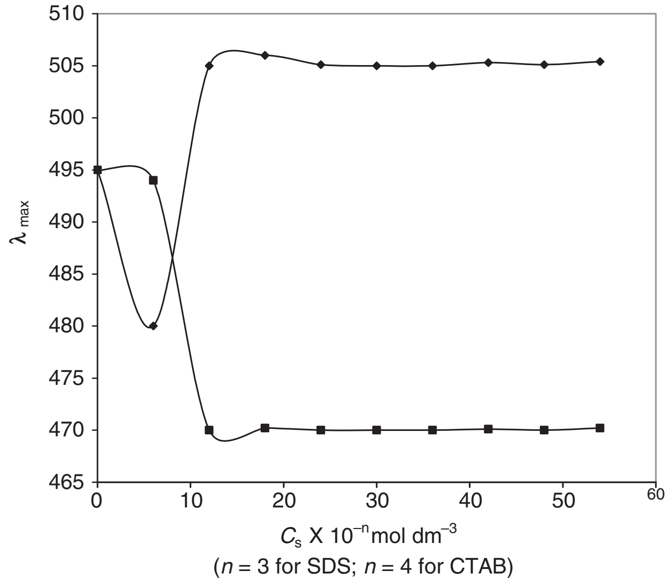


Figure 2. The λ_{max} of NAB as a function of surfactant concentration (◆ SDS, ■ CTAB).

2.2.2. *Spectral shifts*

Electronic spectra of dye, with and without surfactants were recorded on Perkin Elmer (model λ-20) UV-visible spectrophotometer at $25 \pm 0.1°C$. The dye concentration (i.e. 1×10^{-5} mol dm^{-3}) was kept constant throughout the experiment.

2.2.3. *Differential absorbance*

A dye solution of particular concentration (1×10^{-5} mol dm^{-3}) was prepared. This solution was used as solvent for preparation of surfactant solutions of various concentrations. The difference spectra were measured (at 25°C) by setting the cuvette filled with the dye solution in the reference side and one with the surfactant solution at same dye concentration in the sample side of the spectrophotometer. The temperature was controlled within ±0.1°C by using a thermostat.

2.2.4. *Solvent–water partition coefficients*

A solution (1×10^{-5} mol dm^{-3}) of NAB dye in a solvent of choice was kept at 25°C for 1 h. Visible spectrum was then run, and the value of absorbance at the maximum wavelength was measured (A_o). Equal volumes of organic solution and water were mixed. Visible spectra of organic layer were run at regular intervals until the absorbance value became constant (A_x). Due to low solubility of dye in pure hydrocarbons; 5% DMSO solutions were used instead.

3. Results and discussion

The spectral behaviour of NAB dye as a function of surfactant concentration (C_s) is summarised in Figure 2. With a fixed dye concentration, surfactant concentration is varied from values below the critical micelle concentration to level almost eight-folds the cmc. In sub-micellar CTAB, the visible absorption spectra of the dye remain unperturbed. However an abrupt shift of 25 nm to shorter wavelength is observed with broadening of band as CTAB concentration is raised to cmc value. This shows that the formation of surfactant–dye clusters proceeds readily after cmc is reached. These hypsochromic shifts are likely to arise due to the possibility of cationic surfactant to constrain the electron pair present on hydrazo nitrogen of the dye [19]. This happens only after the intramolecular interactions in NAB are overcome in the micellar region [36,37].

In the case of SDS, a blue shift of 15 nm is observed at surfactant concentration below cmc, and then a red shift of 10 nm beyond original value is seen along with spectral broadening as cmc is approached. Such response is ascribed to the growth of pre-micellar aggregates, followed by the transfer of dye within anionic micelles [38].

Figure 3(a) and (b) shows that a significant increase in the absorbance of the band at 418 nm occurs as the surfactant concentration exceeds the cmc. This reflects a tautomeric shift due to change in microenvironment of dye [39]. However, this increase in absorbance is not accompanied by a concurrent decrease in the intensity of absorption maximum representing the corresponding hydrazone form. This could be interpreted as the occupation of two different residence sites by NAB within cationic and anionic micelles.

The formation of dye–surfactant aggregates as well as the distribution of dye between aqueous and micellar phase have been some times described by the association equilibria [16,20,40–44] where

$$iD + jS \rightleftarrows D_iS_j,$$

illustrates the formation of mixed clusters between dye (D) molecules and monomeric surfactant (S) and

$$yD + S_\mathrm{n} \rightleftarrows D_\mathrm{y}S_\mathrm{n},$$

shows the distribution of dye (D) among micelles of normal type S_n [7].

Tokiwa's equation [45] was adapted for evaluating the average number of molecules per one dye molecule (n_S) just above cmc:

$$n_s = \frac{C_\mathrm{s}^{\mathrm{m}_\circ}(\varepsilon_\mathrm{o} - \varepsilon_\mathrm{m})}{A_\circ - A}, \tag{1}$$

where $A_\circ$ and A denote the absorbance of dye in aqueous and micellar phase and $\varepsilon_\circ$ and ε_m refer to the corresponding absorptivities. $C_\mathrm{s}^{\mathrm{m}_\circ}$ is the surfactant concentration less than the critical micelle concentration.

Table 1 reports some experimental and calculated data including dye–surfactant ratio ($n_\mathrm{D}/n_\mathrm{S}$) in mixed micelle. For CTAB, one dye molecule is associated with 10 surfactant molecules in the hybrid clusters. $n_\mathrm{S} = 39$ for SDS and gives $n_\mathrm{D}/n_\mathrm{S} = 0.025$, evident of surfactant rich aggregates.

The equilibrium constant 'K_c' for the distribution of dye between aqueous and micellar phase can be expressed in terms of the mole fraction of AAB in two phases.

$$K_\mathrm{c} = \frac{X_\mathrm{m}}{X_\mathrm{a}} \tag{2}$$

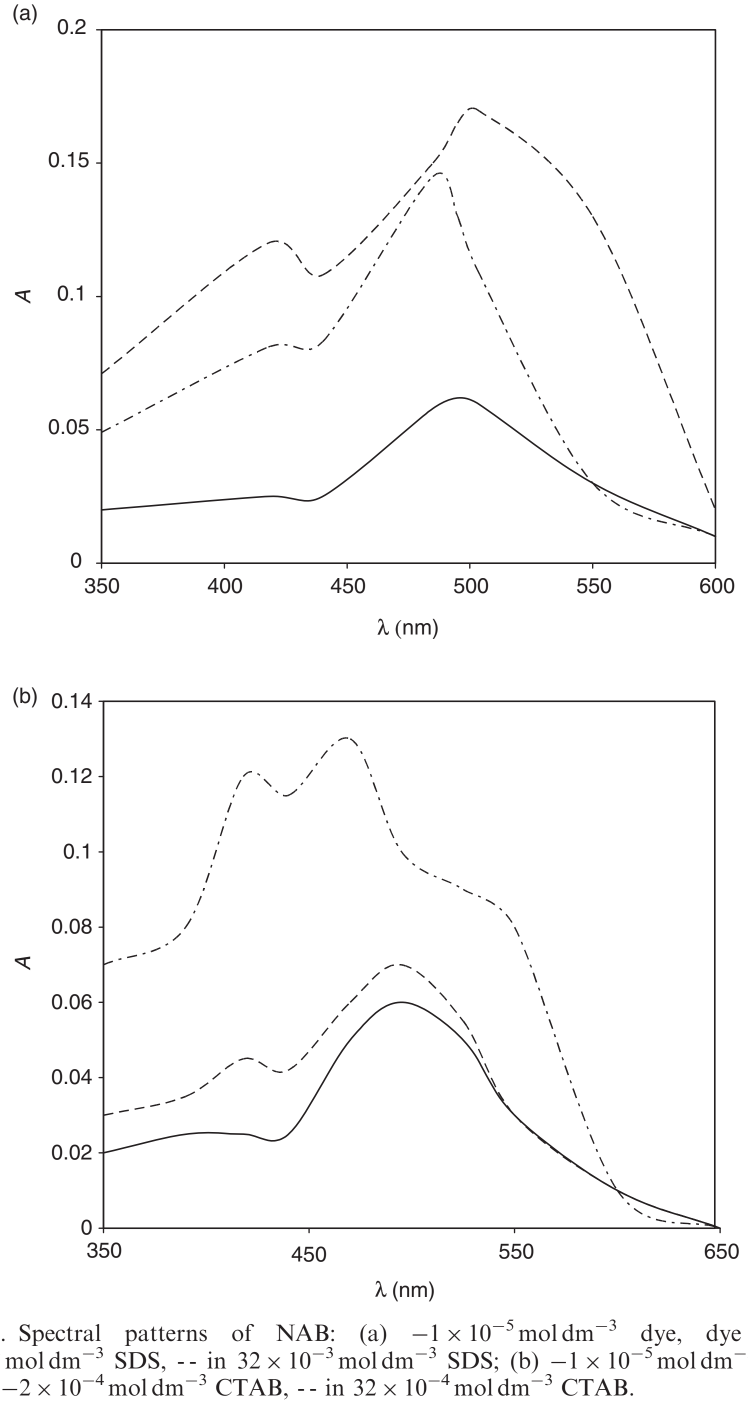


Figure 3. Spectral patterns of NAB: (a) $-1 \times 10^{-5}\,\mathrm{mol\,dm^{-3}}$ dye, dye in SDS $-2 \times 10^{-3}\,\mathrm{mol\,dm^{-3}}$ SDS, -- in $32 \times 10^{-3}\,\mathrm{mol\,dm^{-3}}$ SDS; (b) $-1 \times 10^{-5}\,\mathrm{mol\,dm^{-3}}$ dye, dye in CTAB $-2 \times 10^{-4}\,\mathrm{mol\,dm^{-3}}$ CTAB, -- in $32 \times 10^{-4}\,\mathrm{mol\,dm^{-3}}$ CTAB.

Table 1. Experimental and calculated parameters characterising surfactant–dye association.

Surfactant	$C_s \times 10^{-4}$ (mol dm^{-3})	A_o	A	$\varepsilon_m \times 10^3$ (dm^{3}mol^{-1}cm^{-1})	$C_m \times 10^6$ (mol dm^{-3})	n_s	n_D/n_s
CTAB	10.0	0.09	0.122	12.5	9.14	10	0.10
SDS	85.0	0.10	0.135	14.6	7.60	39	0.025

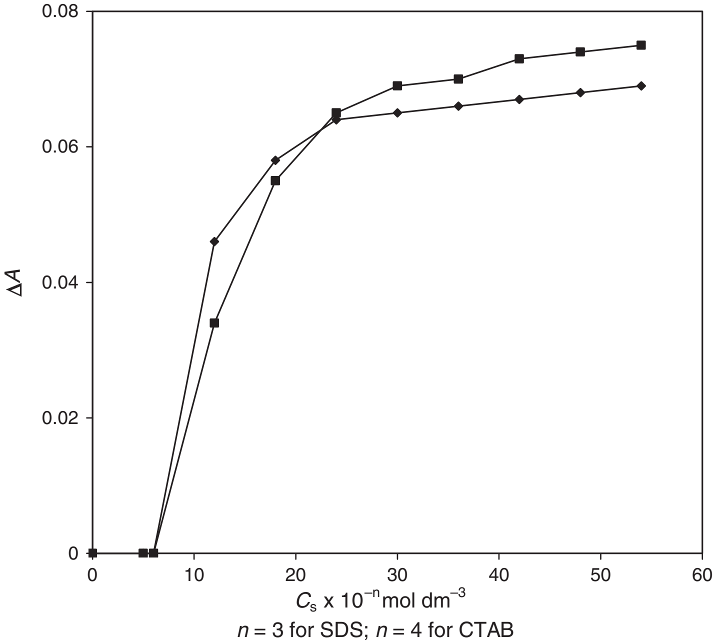


Figure 4. Impact of surfactants on differential absorbance of NAB (◆ SDS, ■ CTAB).

The translation of Equation (2) in linear form produces [46]

$$\frac{1}{\Delta A} = \frac{1}{K_c \Delta A_\infty (C_d + C_s^{m_\circ})} + \frac{1}{\Delta A_\infty}, \tag{3}$$

where C_d is the total dye concentration and $C_s^{m_\circ}$ is the analytical concentration of the micellised surfactant. ΔA_∞ is the differential absorbance at infinite C_s, a concentration where all dye molecules are assumed to have incorporated into the micelles. $\Delta A \approx 0$ for sub-micellar concentrations of two surfactants and increase progressively for all C_s values greater than cmc (Figure 4). This directly indicates the solubilisation of greater proportion of C_d into the micelles. K_c values obtained from the plot of $1/\Delta A$ against $1/(C_d + C_s^{m_\circ})$ are displayed in Table 2 and Figure 5(a) and (b). The partition coefficients yielded for two surfactants differ considerably and confirm that dye is preferentially solubilised within CTAB micelles, earlier indicated by n_D/n_S ratio calculated for SDS–NAB and CTAB–NAB systems. For further investigation regarding the influence of CTAB and SDS,

Table 2. Maximum energy of transition (ΔE_T), partition coefficients (K_c, K_x) and Gibbs free energy (ΔG_p°) for dye–surfactant system.

Surfactant	$E_T \times 10^{19}$ (J)[a]	K_c ($dm^3 mol^{-1}$)	K_x	ΔG_p° ($KJ mol^{-1}$)
CTAB	1.428	1591	88320	−28.16
SDS	4.499	435	24134	−25.00

Note: [a]Obtained using method given in [54].

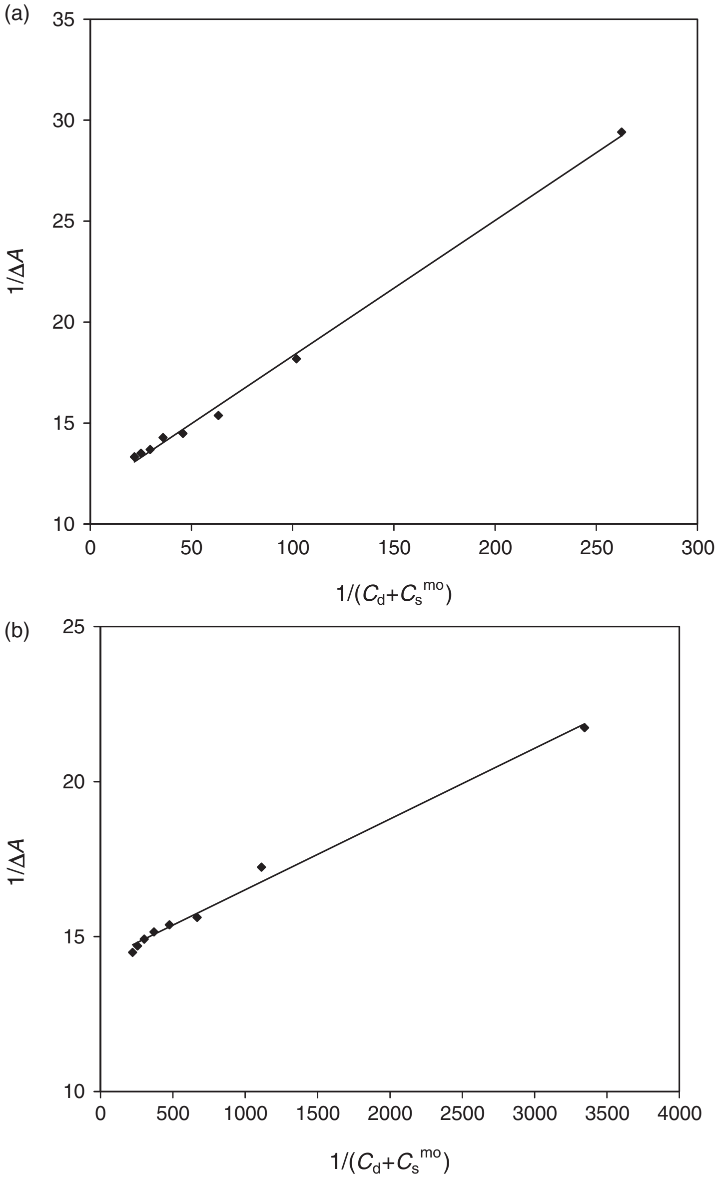


Figure 5. $1/\Delta A$ against $1/(C_d + C_s^{m_o})$ for (a) SDS–NAB system and (b) CTAB-NAB system.

the relative solubility ($S_t/S_\circ$) of NAB was ascertained by employing the following relation [47]:

$$\frac{S_t}{S_\circ} = 1 + K_x \nu M, \tag{4}$$

where S_t and S_o denote total and intrinsic water solubilities.

$$K_x = K_c n_w, \tag{5}$$

n_w is the moles of water per cubic decimeter, i.e. 55.5 M, the micelle is given by

$$M = \frac{C_s - CMC}{N_{agg}}. \tag{6}$$

Here N_{agg} represents the micellar aggregation number. Average values of N_{agg} for SDS and CTAB are reported widely [48] and were used to calculate micellar concentration (M).

The simplified relations [Equations (7) and (8)] were used for estimation of partial molal volumes (ν) SDS and CTAB, utilising reported area per surfactant molecule at surface saturation [49].

$$\nu_{SDS} = \left(\frac{1}{\pi}\right)^{0.5} \beta(A)^{1.5} \tag{7}$$

$$\nu_{CTAB} = \left(\frac{1}{2\pi}\right)^{0.5} \beta(A)^{1.5} \tag{8}$$

$$\beta = \frac{N_A (N_{agg})^{1.5}}{6},$$

where β is an empirical parameter that reflects aggregation tendency of a particular surfactant and its impact on partial molal volume is shown in Equations (7) and (8). The values computed for SDS and CTAB were well in agreement with the reported values, i.e. 0.2464 and 0.3654 $\text{dm}^3\,\text{mol}^{-1}$ [49]. The parameters characterising the relative solubility of dye in the examined ionic surfactants are presented in Table 3. $S_t/S_\circ$ increases upon increasing C_s for both CTAB and SDS. The gap between S_t and $S_\circ$ for one particular surfactant widens with increase in magnitude of M, suggesting enhanced solubility of dye in micellar phase. For same micellar concentration M, the relative solubility of NAB is higher in cationic surfactant CTAB in comparison to SDS.

The Gibbs free energy for transfer of dye molecules from aqueous to micellar phase (ΔG_p^o) can be expressed as

$$\Delta G_p^o = -RT \ln K_x. \tag{9}$$

Again ΔG_p^o is more negative for CTAB and reflects an ease of transfer to cationic micelles. The critical micelle concentrations for ionic surfactants determined conductometrically are comparable to literature values (Table 4). Surface tension studies conducted by Khamis *et al.* [26] shows independence of cmc of SDS and CTAB in the presence of azo dyes, methyl orange and acid alizarin violet N. But the

Table 3. Relative solubility (S_t/S_o) of azo dye at different micellar concentrations.

CTAB			SDS		
$C_s \times 10^{-4}$ (mol dm^{-3})	$M \times 10^5$ (mol dm^{-3})	S_t/S_o	$C_s \times 10^{-4}$ (mol dm^{-3})	$M \times 10^5$ (mol dm^{-3})	S_t/S_o
12.0	0.32	1.032	120.0	5.3	1.315
18.0	0.98	1.316	180.0	13.8	1.820
24.0	1.65	1.532	240.0	22.2	2.320
30.0	2.32	1.748	300.0	30.7	2.825
36.0	2.98	1.961	360.0	39.1	3.325
42.0	3.65	2.177	420.0	47.6	3.830
48.0	4.32	2.394	480.0	56.0	4.330
54.0	4.98	2.607	540.0	64.5	4.835
60.0	5.65	2.823	600.0	72.9	5.335

Table 4. Critical micellar concentration (cmc), interaction coefficient (I_s) and depression in cmc caused by NAB.

	Critical micelle concentration (cmc) $\times 10^{-4}$ (mol dm^{-3})			
Surfactant	Present work	Literature[a]	I_s	$\frac{-\mathrm{d}(CMC)}{\mathrm{d}C_d}$
CTAB	9.11	9.20	0.69[b]	0.897
SDS	0.82	0.82	0.62[c]	2.460

Notes: [a]Taken from [48].
[b]Taken from [51].
[c]Taken from [52].

analysis of NAB–surfactant system carried out in this work confirms that NAB induces the formation of surfactant aggregates. The change in cmc of ionic surfactant caused by NAB was calculated using micelle water partition coefficients determined earlier as [50].

$$\frac{-\mathrm{d}(CMC)}{\mathrm{d}C_\mathrm{d}} = \frac{K_\mathrm{x} I_\mathrm{s} CMC_\mathrm{o}}{n_\mathrm{w}}, \tag{10}$$

where $-\mathrm{d}(CMC)/\mathrm{d}C_\mathrm{d}$ is the depression in cmc and I_s an interaction coefficient of surfactant [51,52].

Though the effect is not considerably large (Table 4), it cannot be ignored in the conditions where solubilisate concentration is high enough to disturb the aggregation process. Only a little effect of the presence of NAB on cmc values can be easily explained keeping in view the low dye concentration used in present study, which is certainly not expected to effect essentially the macroscopic physical property of the system.

In addition to dye structure, the gradient of polarity offered by micelles plays a major role in the distribution of dye between micelle and water phase. The partition coefficients (p) of dye in solvent/water two-phase systems were measured

Table 5. Solvent–water partition coefficients (P) and free energy change for the transfer (ΔG°) of NAB.

Solvent	A_o	A_x	P	$\Delta G^{\circ a}$ (KJ mol^{-1})
n-Pentane	0.131	0.047	0.56	+1.43
n-Hexane	0.129	0.044	0.51	+1.67
n-Heptane	0.136	0.052	0.62	+1.18
1-Butanol	0.294	0.259	7.4	−4.96
1-Hexanol	0.286	0.280	46.66	−9.52
1-Octanol	0.297	0.294	98	−11.36
Carbon tetrachloride	0.158	0.073	0.86	+0.37

Note: $^a\Delta G^\circ = -RT\ln P$.

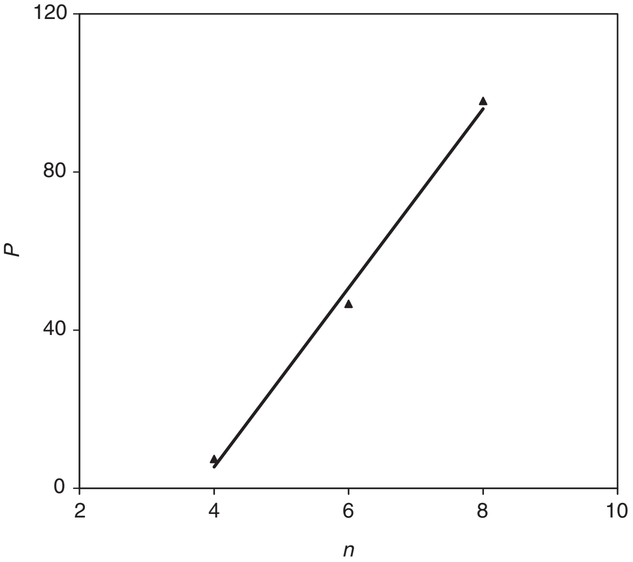


Figure 6. Effect of alkanol hydrophobicity on solvent–water partition coefficient of NAB.

to underline the dye's affinity towards micro polarity of various regions within micelles [53].

$$P = \frac{D_{\text{solvent}}}{D_{\text{water}}} = \frac{A_x}{A_o - A_x}, \tag{11}$$

where D_{solvent} and D_{water} are the concentrations of dye in solvent and water, respectively. A_o and A_x are the initial and final absorbance values of the organic layer.

The solvent water partition coefficients of NAB are delineated in Table 5. All alkane–water partition coefficients are small as well as comparable and the only information that emerges from this data is that hydrocarbon-like micellar interior is impervious to NAB. Among *n*-alkanol–water system, the azo dye NAB showed greater proclivity for higher alcohols. A co-relation between alkanol–water partition coefficient and number of carbon atoms in alkyl chain of organic solvent (alkanol) is illustrated in Figure 6. It is obvious to remark that the aggregation tendency of

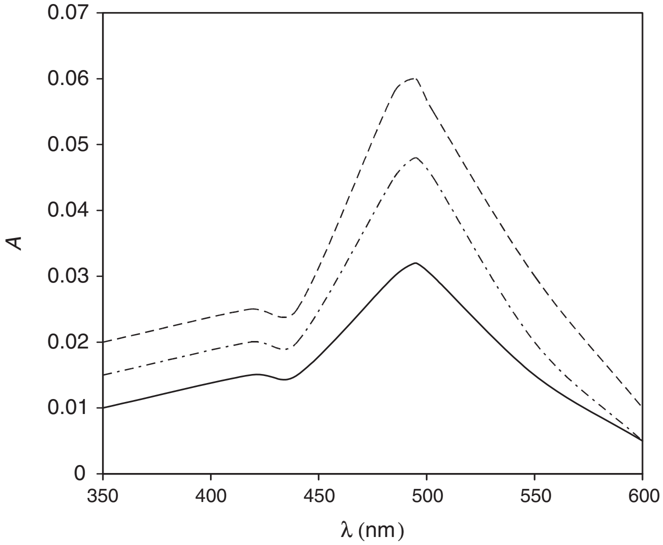


Figure 7. Electronic spectra of NAB at different C_d ($-1\times10^{-6}\,\mathrm{mol\,dm^{-3}}$, $-5\times10^{-6}\,\mathrm{mol\,dm^{-3}}$, $-1\times10^{-5}\,\mathrm{mol\,dm^{-3}}$).

alkanol is responsible for relatively higher *p*-values. The dielectric of 1-butanol is parallel to the dielectric of micelle–water interface of CTAB [54]. As dye is more inclined towards higher alkanol, it may be proposed that NAB is buried slightly deeper within cationic micelles.

The electronic spectra of dye recorded over concentration range (1×10^{-6}–$1\times10^{-5}\,\mathrm{mol\,dm^{-3}}$) remain unchanged (Figure 7). Hence, it may be concluded that NAB does not undergo self-aggregation [4] at the low concentration used in this work. The same applies to the small dependence of cmc in the presence of dye. Initial investigation into the association process reveals the formation of pre-micellar aggregates with anionic surfactant SDS, followed by distribution of dye between aqueous and micellar pseudo phase. In the case of cationic surfactant CTAB, the binding process is indeed cooperative and exhibited, and micelle–water partition coefficient is significantly high for CTAB–NAB system. Detailed analysis of spectral pattern shows the conversion of predominant hydrazone form of dye to corresponding azo form upon transfer of charged micelles. Multiple residence sites for NAB are conceived, based on structural transition, which is nevertheless reversible.

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