Journal of Photochemistry & Photobiology, B: Biology 161 (2016) 59–70

Contents lists available at ScienceDirect

Journal of Photochemistry & Photobiology, B: Biology

journal homepage: www.elsevier.com/locate/jphotobiol

Surfactants induced release of a red emitting dye from the nanocavity of a molecular container: A spectroscopic and calorimetric study

Sayeed Ashique Ahmed, Aninda Chatterjee, Banibrata Maity, Debabrata Seth *

Department of Chemistry, Indian Institute of Technology Patna, Patna 801103, Bihar, India

ARTICLE INFO

Article history:
Received 14 March 2016
Received in revised form 19 April 2016
Accepted 9 May 2016
Available online 11 May 2016

Keywords:
Cucurbit[7]uril
Nile blue A
Supramolecular complex
Ionic liquid
Isothermal titration calorimetry
Release of dye

ABSTRACT

Supramolecular interaction of a red emitting dye Nile blue A (NBA) with Cucurbit[7]uril (CB7) in aqueous solution was studied and the release of the dye from the hydrophobic cavity of CB7 was reported. To investigate the supramolecular host–guest complex formation and release of dye, we have used the steady state absorption, fluorescence and time resolved fluorescence emission spectroscopy, ^{1}H NMR spectroscopy and isothermal titration calorimetry (ITC). The spectral properties of NBA were changed in the presence of CB7. The change in spectral features of NBA in presence of CB7 indicates the formation of supramolecular host–guest complexes. By using the SED equation the diameter of the complex was estimated. The complex formation further affirmed by the ^{1}H NMR study. Upfield and downfield shifts of the protons of NBA was observed in both the aliphatic and aromatic region. From the ITC measurement, we have drawn up the forces involved for the complexation of NBA with CB7. We have studied the release of NBA from the hydrophobic cavity of CB7 by using ionic, neutral surfactants and ionic liquid with the help of spectroscopic and calorimetric techniques. It is observed that on addition of SDS and ionic liquid (<*cmc*) ion-pair formation takes place between NBA and surfactant monomer whereas, it was not observed for neutral and cationic surfactant. Above *cmc* of the surfactants, complex is formed between NBA and micelle.

1. Introduction

The interactions between rigid molecular containers with small molecules are thriving research area for a long time, since it has ubiquitous applications in different fields such as sensors, drug delivery, catalysis, supramolecular dye lasers [1–8]. There are wide ranges of unnatural receptor molecules are available, which are synthesized to enhance the solubility of the guest molecule within the medium and stability of the guest molecule by shielding them from the outside environment. Some examples of important and frequently used receptors are cyclodextrins, calixarenes and cucurbit[*n*]urils [1,9–11]. Recently, several research groups gave more attention to use cucurbit[*n*]urils in many purposes such as drug-delivery, formation of molecular necklaces, etc. as it has higher affinity towards guest molecule [1,4,12–13]. CBn are pumpkin-shaped, where glycoluril units linked by a pair of methylene groups. Cucurbit[7]urils (CB7) is a highly symmetric, rigid macrocyclic host, consists of seven glycoluril units connected to each other by a pair of methylene group in a cyclic manner [14]. On the basis of number of glycoluril unit ($n = 5$–10) CBn have varying cavity diameter and portal sizes. Cucurbit[*n*]urils have nonpolar cavity like other macrocyclic host and also has a polar carbonyl portal end. Hence, hydrophobic interactions as well as strong ion–dipole and charge–dipole interaction due to the presence of carbonyl portal ends with guest molecules are observed [1]. The Coulombic interaction between highly polarisable carbonyl portals of cucurbit[*n*]urils and the host molecule provides exceptionally greater stability to the complex as compared to the other frequently used host molecules like cyclodextrins, calixarenes, etc. It brings cucurbit[*n*]urils in limelight in the field of supramolecular chemistry and diverse their exceptional potentials for various suitable applications. Here, we studied the host–guest complexation of Nile blue A (NBA) and CB7 and the release of NBA from the hydrophobic cavity of host by using different surfactants and an ionic liquid.

NBA is one of the photosensitive, electroactive and a member of the benzophenoxazine class of dye, such class of dye have low toxicity due to quantum yield of $^1O_2$ generation is very small and some of them temporarily inhibit tumour growth [15–18]. NBA can covalently interact with discrete sites of the individual DNA helices, used as biosensors such as used in gel electrophoresis as marker for the DNA detection [19–20]. It is also used as indicator in acid base reaction and as a staining agent for actinomycin in pathological tissues [21]. NBA shows thermochromic and solvatochromic behaviour in visible region [22]. There is few literature on the photophysics of NBA [22–26]. In this work we have studied the supramolecular interaction between a red emitting dye (NBA) and macro-cyclic host (CB7) by using different types of spectroscopic, isothermal titration calorimetry (ITC) techniques. The chromophoric guest molecules on complexation with molecular containers show the absolute change in their photo-physical as

* Corresponding author.
E-mail address: debabrata@iitp.ac.in (D. Seth).

http://dx.doi.org/10.1016/j.jphotobiol.2016.05.009
1011-1344/

well as other properties, which helps to understand the modulation of molecular properties of guest [27]. There are few literatures where the complexation of dye with macrocyclic host are reported along with the sensitive release of guest molecule from bounded state in well controlled manner by using foreign substances [28–33]. Here we have focused on the host–guest complexation along with systematic release of guest molecule from the hydrophobic nanocavity of CB7 by using cationic cetyltrimethylammonium bromide (CTAB) surfactant, anionic surfactant sodium dodecyl sulfate surfactant (SDS) and neutral Triton X-100 surfactant (TX-100) along with 1-butyl-3-methylimidazolium octyl sulfate ionic liquid ([BMIM][OctS]). Surfactants have ubiquitous application in different industrial processes such as waste treatment, emulsifiers for food, pharmaceuticals, catalysis, etc. They are unequivocally studied for mimicking biological cell membranes. Surfactants constitute hydrophobic and hydrophilic groups and from organized assemblies like micelles, vesicles, etc. In recent years the interaction of the surfactants with CB7 has been studied by using different instrumental techniques such as NMR spectroscopic technique, isothermal titration calorimetry (ITC) technique, etc. [34–35]. Inspired by the results of these studies, we are interested to investigate the effect of surfactants on supramolecular host–guest complex where we are presuming the surfactants to be a potential releaser of dye from the hydrophobic cavity of CB7. Again surfactants are biocompatible, biodegradable and having low toxicity for which we choose surfactants as a foreign substance to release dye molecule. This study will be helpful to get the deeper understanding about the interaction of cationic red emitting dye NBA with macrocyclic host CB7. The solubility of NBA in aqueous medium will be increased on complexation with CB7, thereby making it more accessible to the biological site to be stained. Moreover, release of the staining agent NBA using the surfactant will also help us to prudently understand the release mechanism of the dye at the targeted biological site.

2. Materials and Methods

NBA was purchased from Sigma-Aldrich and used without further purification. CB7, CTAB, SDS, TX-100 and [BMIM][OctS] were purchased from Sigma-Aldrich and used as received (Scheme 1). Millipore water was used throughout the experiment.

Ground state absorption measurements were performed with the help of UV–Vis spectrophotometer (Model: UV-2550, Shimadzu). The steady-state fluorescence emission measurements were acquired by using Fluoromax-4P spectrofluorometer (Horiba Jobin Yvon). Absorption and fluorescence measurement were carried out by using quartz cuvette having the path length 1 cm. The fluorescence quantum yields of NBA is different system were measured using the fluorescence quantum yield of NBA in water solution ($\phi_r = 0.004$) as the reference [36] by using the following equation:

$$\phi_f = \phi_r \frac{I_s A_r n_s^2}{I_r A_s n_r^2} \tag{1}$$

where *r* and *s* stand for the reference and sample, respectively. In Eq. (1), *I* stand for the integrated area under the fluorescence curve, *A* stand for the absorbance of the sample at excitation wavelength and *n* stands for the refractive index of the medium. A picosecond time-correlated single-photon counting (TCSPC) technique was used to take the time resolved fluorescence emission decays. We have used a time-resolved fluorescence spectrophotometer from Edinburgh Instruments (model: LifeSpec-II, U.K.). We have used picoseconds diode laser with excitation wavelength at 635 nm. The fluorescence emission decays were taken by using a Hamamatsu MCP PMT (3809U) instrument as the detector at the magic angle (54.7°).

The decays were analysed by using F-900 decay analysis software. The fluorescence anisotropy decays (r(t)) were determined by using the same instrument. To obtain r(t), we used the following equation:

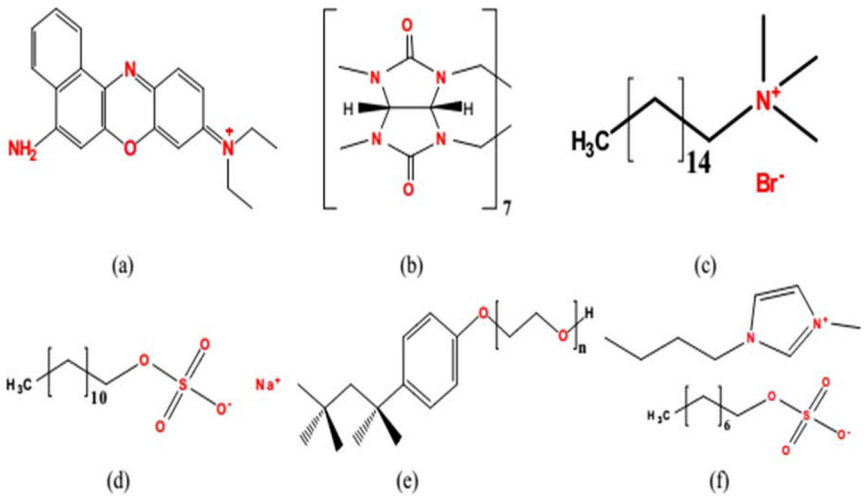
$$r(t) = \frac{I_{II}(t) - G I_{\perp}(t)}{I_{II}(t) + 2G I_{\perp}(t)} \tag{2}$$

where the emission intensities at parallel ($I_{||}$) and perpendicular (I) polarizations were obtained alternatively by fixing the time for both the decays. We have used motorised polarizers to collect the parallel and perpendicular decays. F-900 software was used to analyze the anisotropy decay. During time resolved measurements temperature was kept constant at 298 K by using Peltier-controlled cuvette holders from Quantum Northwest (model: TLC-50). ^{1}H NMR spectral data were collected by JEOL 400 MHz NMR spectrometer. We have used iTC$_{200}$ microcalorimeter from GE healthcare to perform isothermal titration calorimetry measurement and the temperature was kept constant at 298 K during the measurement.

3. Result and Discussion

3.1. Steady State Absorption Measurements

We have performed the steady sate absorption measurement of aqueous solution of NBA in the presence of CB7. We have also studied the effect of cationic (CTAB), anionic (SDS), neutral (TX-100) surfactants as well as [BMIM][OctS] ionic liquid on host–guest complex. We observed that NBA exhibits absorption maximum at 634 nm in water



Scheme 1. Schematic representation of the structure of (a) NBA; (b) CB7; (c) CTAB; (d) SDS; (e) TX-100; (f) [BMIM][OctS].

(Fig. S1, Table S1) and after addition of CB7 to the aqueous solution of NBA, it shows barthochromic shift along with hyperchromic shift (Fig. S1, Table S1). The absorbance maximum of NBA is shifted to 642 nm with increase of relative intensity due to addition of 5.36 μM CB7. Such changes in absorption maximum on addition of CB7 indicate that interaction takes place between NBA and CB7. This interaction arises from the charge–dipole interaction between the cationic form of NBA and polarisable carbonyl portals of CB7 along with the hydrophobic interaction. Further we have studied the effect of addition of different kinds of surfactants and an ionic liquid forming micelles on supramolecular host–guest complex (Fig. 1). On addition of CTAB surfactant (<critical micelle concentration (*cmc*)) to the host–guest complex, sharp hypsochromic shift was observed on absorbance maximum (Fig. 1(a)). It may due to the release of NBA from the hydrophobic cavity of macrocyclic host and with increasing the concentration of CTAB, the absorbance maximum shift towards higher wavelength (639 nm) (Fig. 1(a)). The probable cause for this at higher concentration of CTAB (>*cmc*) is due to NBA attached to hydrophobic part of the micelles. At higher concentration, CTAB (>*cmc*) forms micelle and two types of interaction taken place between micelle and cationic NBA, one is electrostatic interaction and another one hydrophobic interaction. Between these two interactions, hydrophobic interaction is the predominant factor over repulsive electrostatic interaction between cationic head part of the micelle and cationic NBA. Above *cmc*, when micelles are formed the large hydrophobic part of NBA gets solubilised inside the grease like domain of the micelle thereby enhancing the solubility of the dye in the solution. This leaves the cationic part of NBA projected towards the aqueous phase of the solution.

In case of anionic surfactant SDS, on addition of SDS (<*cmc*) a sharp hypochromic shift was observed in absorbance maximum and a new absorbance maximum appeared at 595 nm (Fig. 1(b)). It indicates that new species is generated. The absorbance maximum at 595 nm is due to interaction between the cationic NBA and anionic monomer of surfactant [23]. Such kind of interaction also observed for Rhodamine derivatives and methyl violet [37–38]. This type of phenomenon might also lead to formation of dimmer of the dye [23]. Further increasing the concentration of SDS the absorbance value gradually increased with blue shift. The absorbance maximum observed at 595 nm (<*cmc*) was vanished (Fig. 1(b)). The reason for hyperchromic shift in absorbance on gradual addition of surfactant is due to the complex formation between micelles and cationic dye. The blue shift in absorbance spectra observed due to the cationic dye resides in hydrophobic core of micelles. On addition of neutral surfactant (TX-100), a little changed was found in absorbance spectra (Fig. 1(c)). It indicates that TX-100 has lesser effect on supramolecular host–guest complex as compared to other surfactants used in this work.

We have also studied the effect of addition of 1-butyl-3-methylimidazolium octyl sulfate ([BMIM][OctS]) ionic liquid on host–guest complex (Fig. 1(d)). We have observed similar types of phenomenon as that observed due to addition of anionic surfactant, SDS. Hypochromic shift was observed in absorbance maximum on addition of [BMIM][OctS] (<*cmc*) to the solution of NBA–CB7 complex (Fig. 1(d)). Along with hypochromic shift, a new absorbance maximum was observed at around 600 nm, which implied that a new complex is formed. This new complex may be due to the interaction between cationic dye and anionic part of ionic liquid i.e. premicellar complex formation. Further on increasing the concentration of ionic liquid, the maximum present at 600 nm totally vanished and showed only a single absorbance maximum at 642 nm (Fig. 1(d)). It may be due to the formation of the micelle–NBA complex. It is reported in literature that [BMIM][OctS] forms micelle [39–40]. So, it may be possible that the cationic NBA stacked at the hydrophobic core of micelles (formed by octyl sulfate part of [BMIM][OctS]), for this reason the absorbance showed the hyperchromic shift along with barthochromic shift.

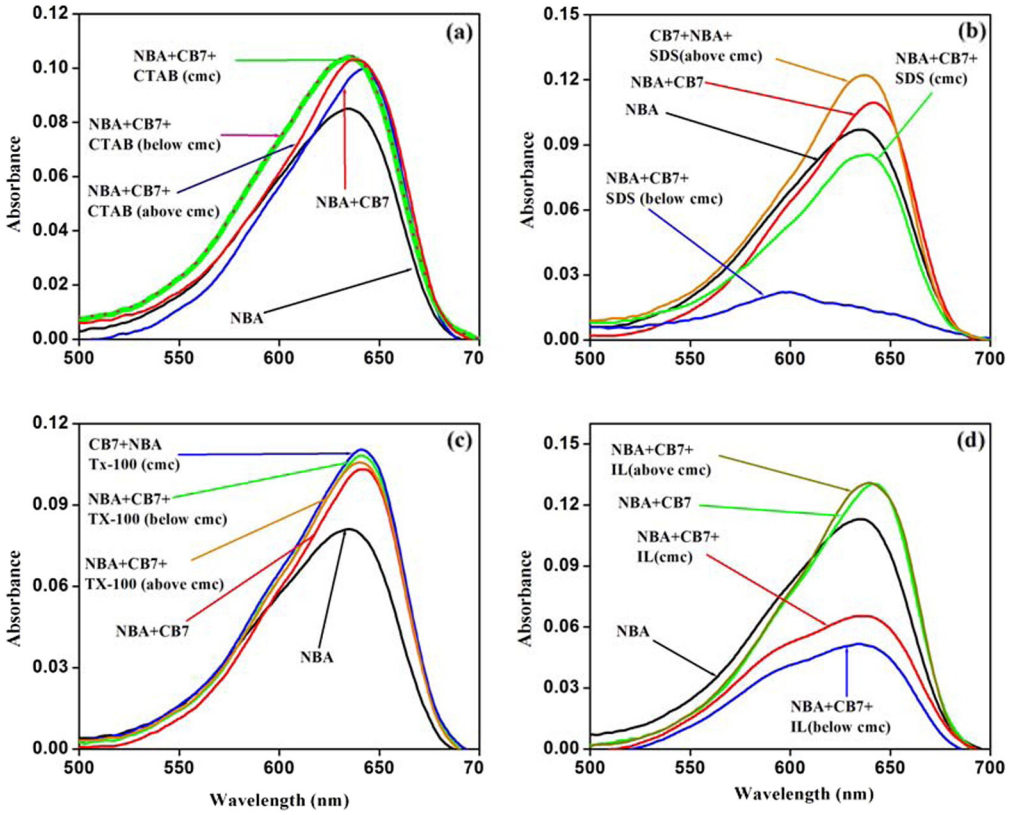


Fig. 1. Absorption spectra of NBA–CB7 complex in water in presence of (a) CTAB; (b) SDS; (c) TX-100; (d) 1-butyl-3-methylimidazolium octyl sulfate [BMIM][OctS].

3.2. Fluorescence Emission Measurements

The steady state fluorescence emission spectrum of NBA in aqueous solution shows single emission maximum at around 672 nm. On addition of CB7 to the aqueous solution of NBA, considerable changes were observed in fluorescence intensity without any change in fluorescence maximum position due to addition of CB7 (Fig. S2, Table S1). We observed fluorescence quantum yield gradually increases with increasing the concentration of CB7 and it is almost double on addition of 5.36 μM CB7 in the aqueous solution of NBA (Table S1). The increase of quantum yield indicates that NBA is facing more restricted environment in presence of CB7 as compared to that of free dye. Such change in fluorescence emission spectra as well as change in quantum yield indicates the formation of NBA–CB7 complex. The reason for the spectral change on addition of CB7 is the charge dipole interaction of positively charged $-NEt_2$ group of NBA with carbonyl portal end of CB7 along with the hydrophobic interaction between NBA and CB7. It was observed that host–guest complexation process completed at very low concentration of CB7. It indicates that CB7 has very strong affinity towards NBA to form the complex.

We have further investigated the effect of different surfactants and an ionic liquid on NBA–CB7 complex. Firstly, on addition of cationic surfactant, CTAB (<*cmc*) to the solution containing complex, it was observed that fluorescence intensity decreased and the fluorescence spectra having the similar type of feature to that of aqueous NBA solution (Fig. 2(a)). It may be possible that NBA released from the hydrophobic cavity of CB7. Again on increasing the concentration of CTAB fluorescence intensity gradually increases. Above *cmc*, CTAB starts to aggregate to form micelles, under such condition the free dye forms complex with micelles. The probable reasons for increase of fluorescence intensity is due to retardation of reversible proton transfer process as compared to that in the bulk water (NBA gets solubilised inside the grease like domain of the micelle) hence, decrease in non-radiative decay rate constant [41–42].

On other hand on addition of SDS (<*cmc*) to the solution containing NBA–CB7 complex, intensity decreased significantly (no emission maximum was observed), it indicates a significant quenching due to ion pair formation between positively charged NBA and SDS monomer [23,37–38]. On further addition of SDS surfactant (>*cmc*) the fluorescence intensity gradually increased along with blue shift (6 nm) (Fig. 2(b)). It indicates that the dye is present in more restricted environment, where the proton transfer process is hindered relative to that of bulk water and the increase of fluorescence intensity along with blue shift, indicates that the complex is formed between cationic dye and micelles and the dye is present at hydrophobic part of micelles [23]. On addition of TX-100 to the aqueous solution of NBA small change was observed in fluorescence spectra unlike ionic surfactants and ionic liquid (Fig. 2(c)). Slight increase in fluorescence intensity was observed. It indicates the formation of the complex between NBA and micelle. Hence, TX-100 has smaller effect on host–guest complex as compared to the others.

On addition of [BMIM][OctS] ionic liquid (<*cmc*) to the solution containing NBA–CB7 complex, it was observed that the fluorescence intensity quenched significantly (Fig. 2(d)). It may be due to the complex formation between monomer of octyl sulfate part of ionic liquid and cationic NBA [23,37–38]. Further increasing the concentration of the ionic liquid (>*cmc*) within the solution, the fluorescence intensity gradually increases along with blue shift (3 nm) (Fig. 2(d)). This change in fluorescence spectra may be due to the interaction between micelle with NBA. Due to this micelle-dye complex formation proton transfer process in NBA is hindered compared to bulk water and the fluorescence

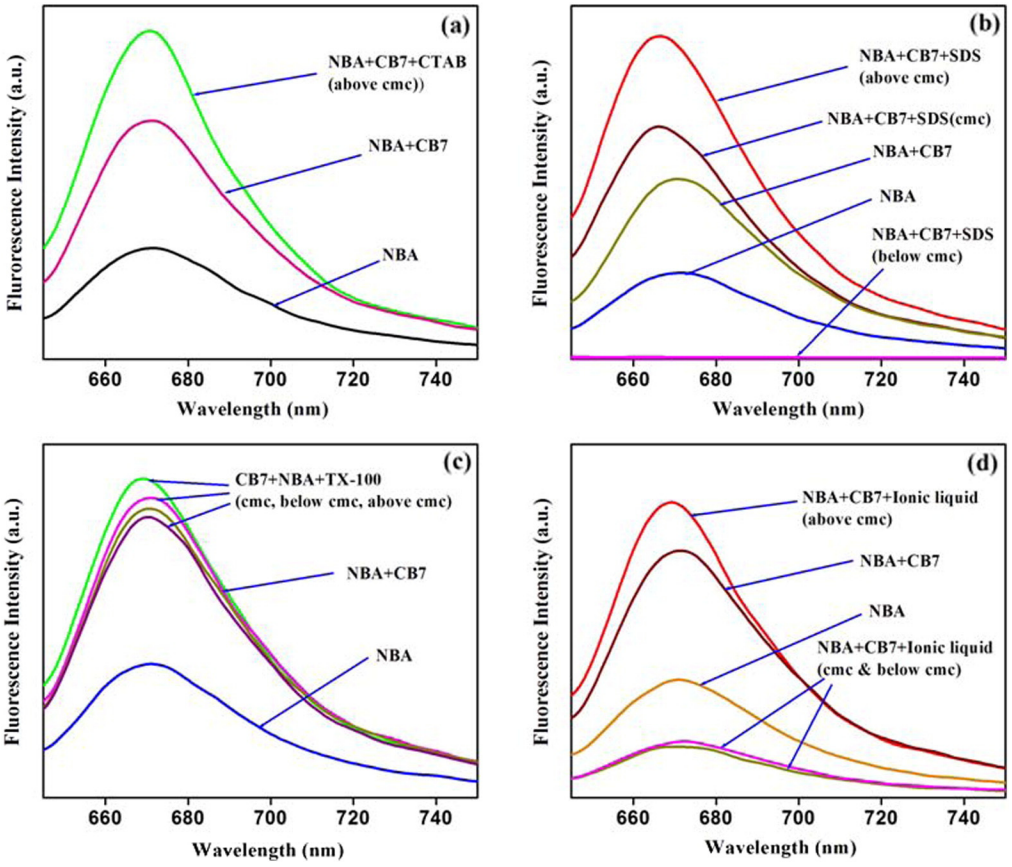


Fig. 2. Fluorescence spectra of aqueous solution of NBA and NBA–CB7 complex and changes observed due to addition of different surfactants, where (a) CTAB; (b) SDS; (c) TX-100; (d) [BMIM][OctS].

intensity increases on gradual addition of [BMIM][OctS] and the blue shift of fluorescence maxima indicates that NBA probably present at nonpolar part of the micelles [41–42].

3.3. Determination of the Binding Constants and Stoichiometry

In our study we found that 1:1 complex is formed. The 1:1 guest–host stoichiometry can be written as follows:

$$NBA + CB7 \leftrightarrow NBA \cdot CB7.$$

The binding constant value (K) of the complex is found by using the following equation, where experimental absorbance data are directly fitted by Eq. (3) [43].

$$A_{obs} = A_0 - \frac{\Delta\varepsilon}{2}\left\{[G]_0 + [CB7]_0 + 1/K - \sqrt{\left\{([G]_0 + [CB7]_0 + 1/K)^2 - 4[G]_0[CB7]_0\right\}}\right\} \quad (3)$$

where, A_{obs} and A_0 are the observed absorbance and absorbance in absence of host. [G] is the concentration of NBA and [CB7] is the concentration of the CB7. The binding constant value for 1:1 complex is found to be 5.8 ($\pm$5.1) $\times$ 10^7 M^{-1} with standard deviation $\sigma(A)$ 0.0002 by using the above Eq. (3) (Fig. S3).

The binding constant value of the complex was estimated from the fluorescence measurement by using nonlinear least-squares regression analysis in which data are directly fitted by using the relevant equation. The initial value of the unknown parameter was obtained with the help of linear fitted result by using Benesi–Hildebrand equation [44].

In our study only 1:1 complex is formed between NBA and CB7. The binding constant value (K_1) is obtained using the following equation

$$F = \frac{F_{water} + F_m K_1[CB7]}{1 + K_1[CB7]} \quad (4)$$

where F_{water} and F_m are the fluorescence intensities in absence and in presence of maximum concentrations of CB7 where 1:1 binding has been completed, respectively. F represents the fluorescence intensities at different concentrations of CB7 and [CB7] represents concentration of CB7. By using the value of fluorescence data, the binding constant value for the 1:1 complex is found to be 6.2 ($\pm$1.9) $\times$ 10^5 M^{-1} (Fig. S4). From binding constant value, it is clear that NBA has higher affinity towards CB7 in ground state as compared to excited state.

3.4. Determination of Stoichiometry of the Complex by Job's Plot

The stoichiometry of the host–guest complex between NBA and CB7 is confirmed by using the Job's method of continuous variation [45]. We are able to determine the stoichiometry by using both absorbance as well as fluorescence data. The Job's method is performed by fixing the total concentration of the solution ([NBA] + [CB7]) at 1×10^{-5} M for both measurements. From Job's plot by using absorbance data, we observed that 1:1 complex formed between NBA and CB7, here we plot $\Delta A \cdot X_{NBA}$ {(difference of absorbance) $\times$ (the mole fraction of the dye)} against X_{NBA} (the mole fraction of the dye) (Fig. S5(a)). From fluorescence measurement it was observed that 1:1 complex is formed between NBA and CB7, here we plot $\Delta F \cdot X_{NBA}$ {(difference of fluorescence intensity) $\times$ (the mole fraction of the dye)} against X_{NBA} (the mole fraction of the dye) (Fig. S5(b)).

3.5. Time Resolved Emission Measurements

The fluorescence decay patterns of NBA in aqueous medium at different concentrations of CB7 are shown in Fig. 3(a). It was observed from lifetime measurement that NBA shows a single exponential decay with time constant 0.35 ns in aqueous medium, which is similar to earlier studies (Table S2) [23,26]. In comparing with the literature, it is observed that NBA has shorter lifetime in polar aqueous medium as compared to non-polar solvent. Shorter fluorescence lifetime in aqueous medium is due to the ultrafast proton transfer process between NBA and water molecule [22,41]. On addition of CB7 to the aqueous solution of dye, spectral feature was changed with increasing average life time; it indicates that complexation processes takes place between NBA and CB7. It was also found from lifetime measurement that on gradual addition of CB7 to the aqueous solution of the dye, the weight percentage of fast component (free dye) gradually decreases (the component remains almost constant in time scale) and a new component (due to the complex formation) was found with 0.73 ns (Table S2). This new slow component is due to the 1:1 complex formations between dye and CB7.

The weight percentage of slow component gradually increases on gradual addition of CB7. On addition of CB7 (5.36 μM), the fluorescence emission decays becomes biexponential in nature, fast component is due to free dye with time constant 0.25 ns (10%) and slow component is due to 1:1 complex formation with time constant 0.73 ns (90%). The significant enhancement of fluorescence lifetime on addition of CB7, clearly demonstrates that the dye molecule present in restricted environment and due to this reason proton transfer process is hindered. From this study it is clearly demonstrated that complex is formed between dye and CB7.

The release protocols of dye molecule from host in the presence of different surfactants and [BMIM][OctS], have been studied by fluorescence emission decay measurements (Fig. 3, Table 1). We have found clear evidence about the release protocol for the trapped dye from host by using fluorescence lifetime measurement. It was found that on addition of CTAB ($<cmc$), the fluorescence life time value decreased to 0.37 ns from 0.68 ns (Fig. 3(b)). The lifetime value free dye molecule in aqueous medium is 0.35 ns. Therefore, it indicates that all dye molecules released from hosts. It is also observed that CTAB shows greater propensity to expel the bound dye molecule from CB7 to aqueous medium as compared to other studied substances used for release. On successive addition of CTAB, the fluorescence life time value increased to 1.09 ns in presence of 76 mM CTAB (Fig. 3(b), Table 1) and we also observed that the fluorescence life time value of NBA increased to 1.10 ns on addition of 76 mM CTAB in the aqueous solution of NBA. Hence, we can say that the component with fluorescence life time value 1.09 ns is due to micelle–NBA complex formation. In earlier sections, we have mentioned that the absorbance maximum show blue shift and fluorescence intensity increase on addition of CTAB ($\geq cmc$) is indicative that NBA probably resides at the hydrophobic core of the micelles. For which the fluorescence life time value increased as compared to the aqueous medium. Again increasing the fluorescence lifetime on addition of 76 mM CTAB is indicated that the proton transfer process is quenched as compared to that observed in the bulk aqueous medium. Initially, addition of SDS ($<cmc$) to the aqueous solution of NBA–CB7 complex, we are unable to measure the life time value as it shows no emission maximum, due to the formation of pre-micellar aggregate with NBA (ion-pair interaction between surfactant and NBA). Such kind of pre-micellar aggregate of surfactant with dye has been reported earlier [38]. However, further addition of SDS ($\geq cmc$) to the aqueous solution of the complex, the fluorescence life time value gradually increases. The average fluorescence life time value increased to 1.12 ns in presence of 69 mM SDS with components 0.68 ns (5%) and 1.14 ns (95%) (Fig. 3(c), Table 1). The fluorescence life time decay of NBA in SDS micelles (69 mM) is single exponential in nature and the lifetime value is 1.14 ns. So, it is concluded from above observations that the component with fluorescence lifetime value 1.14 ns arises due to micelle-NBA complex formation and the component with fluorescence life time value with 0.68 ns may be due to the existence of small percentage of NBA–CB7 complex (Table 1). In earlier sections, we report that blue shift was observed in absorption spectra with increasing

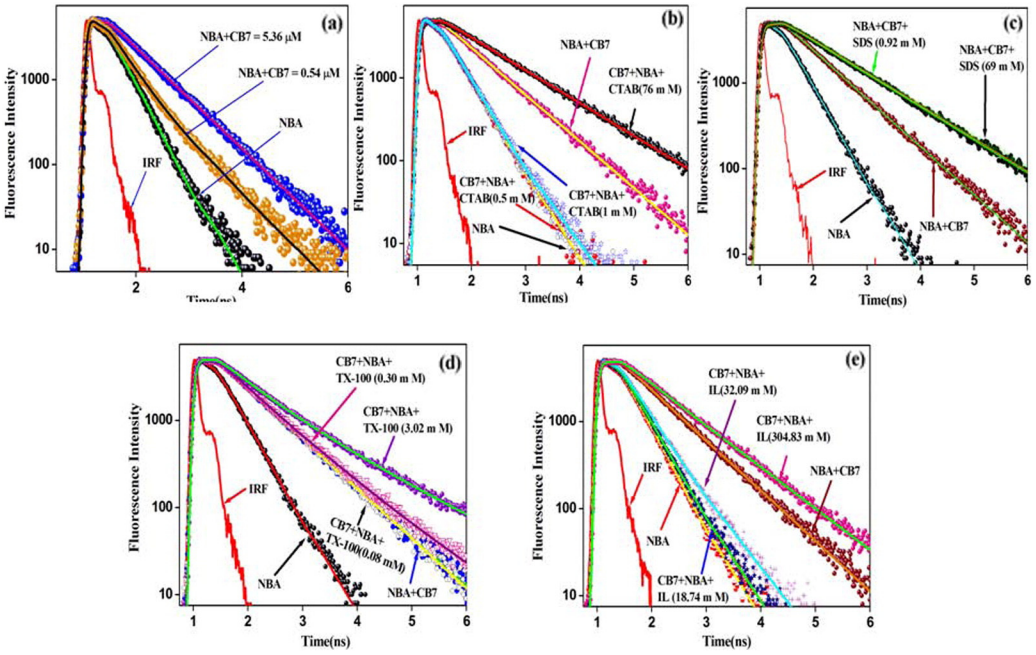


Fig. 3. The fluorescence emission decays of aqueous solution of NBA and in presence of (a) CB7 and with varying the concentration of (b) CTAB; (c) SDS; (d) TX-100; (e) [BMIM][OctS].

fluorescence intensity and in this section lifetime value increases on gradual addition of SDS (>*cmc*). It is concluded from this observation that the complex is formed between micelle and NBA.

The effect of non-ionic surfactant TX-100 is smaller as compared to other studied surfactants and ionic liquid for the release of the dye from the host. On addition of TX-100 (<*cmc*), some dye molecules release but greater amount of dye molecule held together with CB7 (Fig. 3(d), Table 1). It was also observed that on gradual addition of TX-100 to the aqueous solution of the complex the weight percentage of complex molecule decreases and the average life time increases as the formation of complex between micelle and the dye molecule (Fig. 3(d), Table 1). On addition of 3.0 mM TX-100 to the solution of NBA and CB7 the average life time increased to 0.92 ns with components 0.68 ns (66%) and 1.39 ns (34%). The average life time value of NBA in presence of TX-100 micelle (3.0 mM) is found to be 1.01 ns with component 0.36 ns (34%) and 1.34 ns (66%). Hence, we can say that the fluorescence life time component with 1.39 ns may be due to the micelle–NBA complex and fluorescence life time component with 0.68 ns may be due to the existence of NBA–CB7 complex.

Table 1
Effect of addition of surfactants and ionic liquid on host–guest complex between NBA and CB7 on fluorescence emission decay of NBA (λ_{exi} = 635 nm), concentration of NBA is 1.19 μM and CB7 is 5.36 μM.

Sr. no.	System	τ_1 (ns)	a_1	τ_2 (ns)	a_2	τ^c (ns)	χ^2
1	NBA	0.35	1	–	–	0.35	1.041
2	NBA + CB7	0.25	0.10	0.73	0.90	0.68	1.129
3	NBA + CB7 + CTAB (0.05 mM)	0.37	1	–	–	0.37	1.011
4	NBA + CB7 + CTAB (1 mM)	0.35	0.89	0.55	0.11	0.37	0.989
5	NBA + CB7 + CTAB (76 mM)	1.09	1	–	–	1.09	1.028
6	NBA + CTAB (76 mM)	1.10	1	–	–	1.10	1.070
7	NBA + CB7 + SDS (3.95 mM)	–	–	–	–	–	–
8	NBA + CB7 + SDS (8.92 mM)	0.34	0.04	1.12	0.96	1.09	0.961
9	NBA + CB7 + SDS (69 mM)	0.68	0.05	1.14	0.95	1.12	0.969
10	NBA + SDS (69 mM)	1.14	1	–	–	1.14	1.141
11	NBA + CB7 + TX-100 (0.08 mM)	0.29	0.08	0.76	0.92	0.72	1.048
12	NBA + CB7 + TX-100 (0.30 mM)	0.66	0.76	1.06	0.24	0.76	0.984
13	NBA + CB7 + TX-100 (3.0 mM)	0.68	0.66	1.39	0.34	0.92	1.031
14	NBA + TX-100 (3.0 mM)	0.36	0.34	1.34	0.66	1.01	1.023
15	NBA + CB7 + [BMIM][OctS] (18.74 mM)	0.29	0.36	0.44	0.64	0.39	1.119
16	NBA + CB7 + [BMIM][OctS] (32.09 mM)	0.31	0.57	0.53	0.43	0.41	1.101
17	NBA + CB7 + [BMIM][OctS] (304.83 mM)	0.49	0.09	0.91	0.91	0.87	0.920
18	NBA + [BMIM][OctS] (304.83 mM)	0.92	1	–	–	0.92	1.024

$\tau^c = a_1 * \tau_1 + a_2 * \tau_2$.

On addition of [BMIM][OctS] in the solution containing NBA–CB7 complex, similar feature was observed. In the presence of [BMIM][OctS] (<*cmc*) fluorescence emission decays of NBA are fitted bi-exponentially, with time constant 0.29 ns (36%) and 0.44 ns (64%) (Fig. 3(e), Table 1). It indicates that dye molecules were released from the cavity of CB7. Another interesting feature was observed that the generation of a new component with time constant (0.44 ns), it may be due to the formation of premicellar complex between [BMIM][OctS] and NBA. Like SDS, on gradual addition of [BMIM][OctS] the average life time value of NBA gradually increases and on addition of 304.83 mM [BMIM][OctS] average life time value become 0.87 ns (Fig. 3(e), Table 1). We have performed the lifetime measurement of NBA in the aqueous solution of 304.83 mM [BMIM][OctS] and observed single component with time constant 0.92 ns, it may arise due to the micelle–NBA complex formation. Hence, we can say that the fluorescence lifetime component with 0.91 ns (91%) may be due to the micelle-NBA complex formation and existence of small amount (9%) of NBA–CB7 complex.

3.6. Time Resolved Anisotropy Measurements

We have studied the supramolecular host–guest complexation by using the time resolved anisotropy measurement. It was reported that the rotational correlation time of NBA in water is ~0.16 ns [23,26].Whereas, on addition of 5.36 μM CB7 to the aqueous solution of NBA, the rotational correlation time increased to 0.42 ns (Table 2, Fig. 4). It is clearly demonstrated that the complex is formed between NBA and CB7. Due to the complex formation, NBA faces more restricted environment i.e. the rotational relaxation of the dye within CB7 gets highly restricted.

Table 2
The rotational relaxation time of NBA in presence of CB7 and on addition of CTAB, SDS and [BMIM][OctS]; and rotational relaxation time of NBA in presence of CTAB, SDS, TX-100 and [BMIM][OctS].

Sr no	System	r_0	τ_1 (ns)	a_1	τ_2 (ns)	a_2	τ_{rot} [d] (ns)
1.	NBA + CB7	0.298	0.42	1	–	–	0.42
2.	NBA + CB7 + CTAB (76 mM)	0.290	0.58	0.30	3.62	0.70	2.71
3.	NBA + CTAB (76 mM)	0.24	0.76	0.30	3.57	0.70	2.73
4.	NBA + CB7 + SDS (69 mM)	0.309	0.06	0.03	0.97	0.97	0.95
5.	NBA + SDS (69 mM)	0.329	0.07	0.08	1.01	0.92	0.99
6.	NBA + TX-100 (3.0 mM)	0.276	0.13	0.01	3.16	0.99	3.13
7.	NBA + CB7 + [BMIM][OctS] (304.83 mM)	0.242	0.89	1	–	–	0.89
8.	NBA + [BMIM][OctS] (304.83 mM)	0.281	0.88	1	–	–	0.88

τ_{rot} [d] $= a_1 * \tau_1 + a_2 * \tau_2$.

The rotational correlation time increases ~2.5 times as compared to the free dye due to greater hydrodynamic diameter of the host–guest complex compared to that of free dye. We have estimated the value of hydrodynamic radius of the complex by using SED equation [46,47].

$$\tau = \frac{4\pi\eta r_h^3}{3kT} \quad (5)$$

where r_h and η are the hydrodynamic radius of the dye molecule or the host–guest complexes, viscosity of the medium, k is the Boltzmann constant and T is 298 K. The estimated diameter of the dye is found to be ~8.2 Å by using Edward's method [48]. CB7 has outer diameter ~ 5.8 Å, inner diameter ~ 3.9 Å and height ~ 9.1 Å [1]. The estimated radius of the complex is found to be ~7.45 Å from the Eq. (5). Hence, the estimated diameter of the complex is found to be ~14.9 Å. It clearly highlights that the diameter of the rotating species corresponding to this component is lower than the sum of the heights of the CB7 and the diameter of the free dye. It reinforces the concept that the $-NEt_2$ terminals of the dye are residing inside the hydrophobic cavities of CB7 forming 1:1 dye–macrocyclic supramolecular complex (NBA–CB7). The most probable structure of the supramolecular host–guest complex is shown in Scheme 2.

We have also studied the release of the dye molecule from the hydrophobic nano-cavity of CB7 by using the surfactants and ionic liquid with the help of anisotropy measurement. It was observed that on addition of 76 mM CTAB surfactant the average rotational relaxation time increases to 2.71 ns (Table 2, Fig. 4(a)). The average rotational relaxation time of NBA in CTAB micelles (76 mM) was found to be 2.73 ns. It indicates that with the addition of CTAB, NBA molecule release from the cavity of CB7 and interacts with micelles formed by CTAB. With the addition SDS (69 mM) in NBA–CB7 complex the average rotational relaxation time (τ_{rot}) is increased to 0.95 ns compared to that in the presence of CB7 (Table 2, Fig. 4(b)). The average rotational relaxation time of NBA in 69 mM SDS is 0.99 ns. On addition of 3 mM TX-100 in the solution containing NBA–CB7 complex, as it shows "dip-rise-dip" kind of anisotropy decay profile (Fig. S6). The average rotational relaxation time of NBA in the aqueous solution of 3 mM TX-100, was found to be 3.13 ns and the decay is biexponential in nature (Table 2). On addition of 304.83 mM [BMIM][OctS] in the solution containing NBA–CB7 complex, the rotational correlation time increased to 0.89 ns (Table 2, Fig. 4(c)) and the rotational correlation time for NBA in the aqueous solution of 304.83 mM [BMIM][OctS] was found to be 0.88 ns. Hence, it may be due to the formation of the dye–micelle complex. The increase of rotational correlation time indicated that NBA faces more restricted environment in micelles as compared to macrocyclic host.

3.7. ^{1}H NMR Studies of the Host Guest Complexation

The host–guest complexation between NBA and CB7 has been also studied by using the ^{1}H NMR spectroscopic technique (Fig. 5). ^{1}H NMR of NBA with CB7 has been studied in D_2O solvent (Fig. 5). In D_2O the methyl and methylene protons of the $-NEt_2$ group are found at

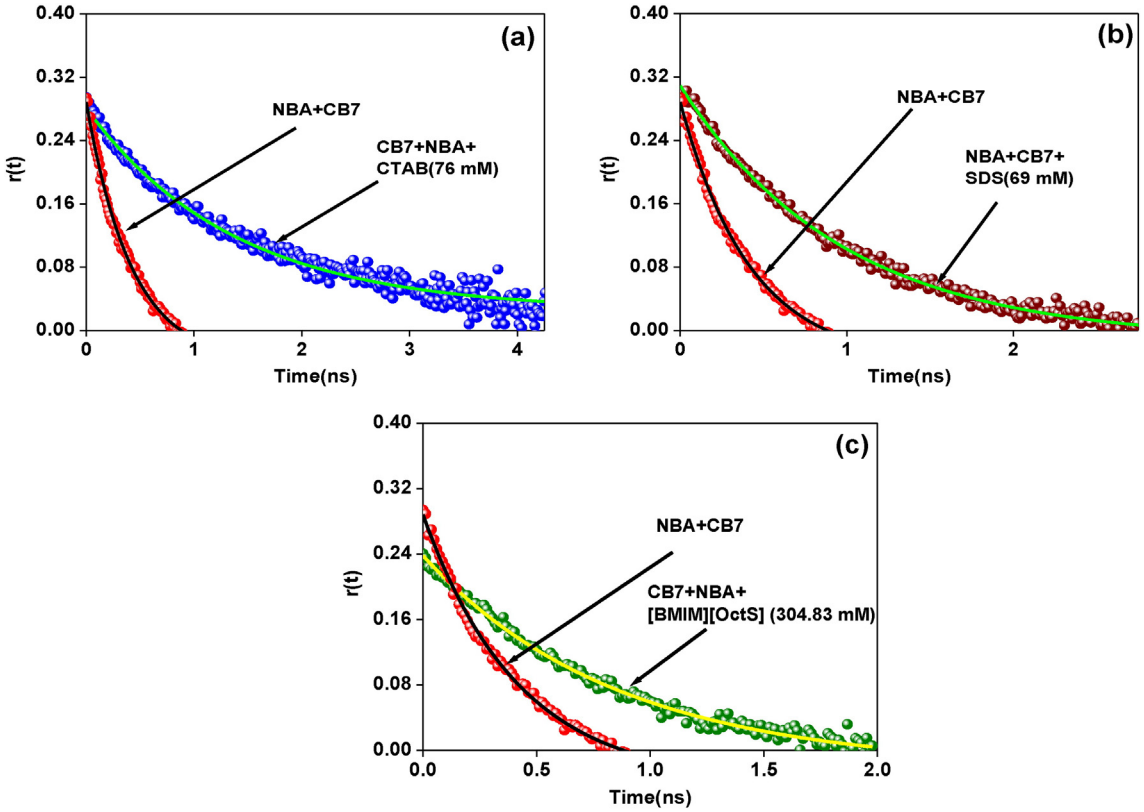
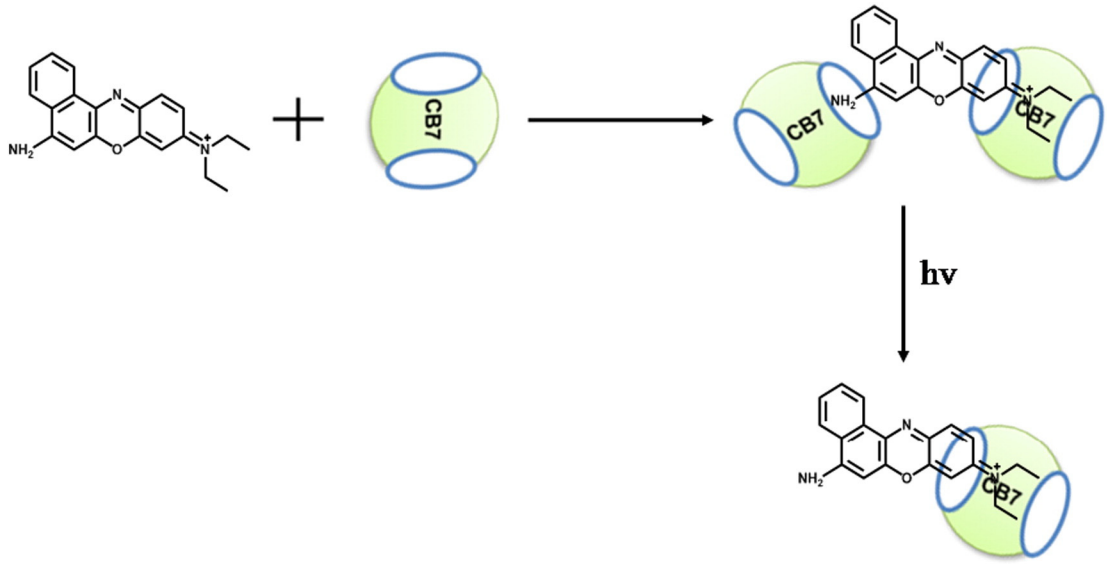


Fig. 4. The time resolved anisotropy decays of NBA in presence of CB7 and after the addition of (a) CTAB; (b) SDS; (c) [BMIM][OctS].



Scheme 2. Schematic representation of probable structure of the complex formation between NBA and CB7.

$\delta = 1.13$ and 3.42, respectively. On addition of CB7 to the solution of NBA, methyl and methylene protons of the $-NEt_2$ group are appeared at $\delta = 0.54$ and 2.90, respectively. Such 0.59 and 0.52 upfield shifts of δ value in NMR spectra clearly indicate the complete inclusion of ($-NEt_2$) part of the dye inside the hydrophobic cavity of CB7. The proton 'c' also shows 0.12 ppm upfield shift, which indicates that, proton 'c' present inside the cavity of the CB7. However, the other protons (d, e, f, g and h) show the downfield shift which may implied that these proton resides near the carbonyl portal of CB7, which also demonstrates that ($-NH_2$) group also interact with the carbonyl portal end for this association of host and guest, proton (f, g and h) show the downfield shift. Whereas, the proton (i and j) do not show observable change in ^1H NMR spectra in presence and absence of CB7 as these protons present far away from the carbonyl portal end of CB7. Hence, ^1H NMR study showed that 1:2 complex is formed between NBA and CB7.

3.8. Study of NBA/CB7 Association and Dissociation Using ITC Measurements

The change of heat on host–guest complex formation can be measured directly by using isothermal titration calorimetry (ITC) titration at temperature 298 K. The heat released or absorbed in the sample cell due to the formation or dissociation of host–guest complex is measured with respect to a reference cell filled with water. With the help of this calorimetric technique, binding nature of the complex, binding

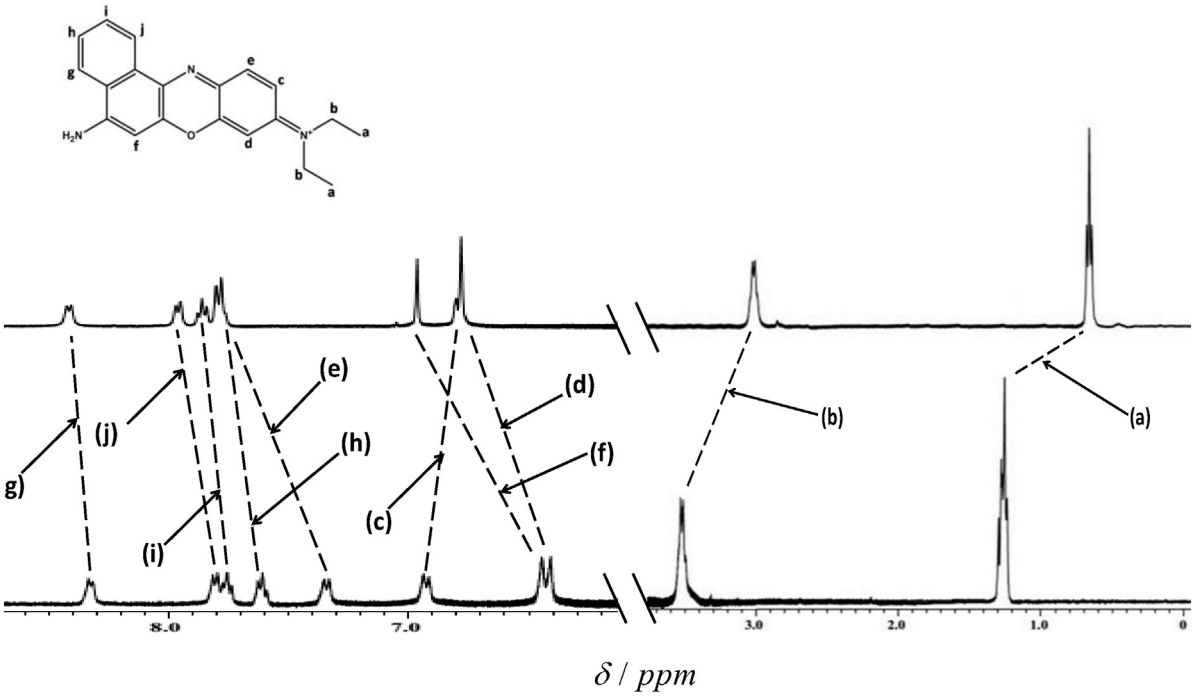


Fig. 5. ^1H NMR spectra (400 MHz) of NBA in D_2O free dye (lower part) and in presence of CB7 (upper part).

enthalpies, binding entropies and Gibbs free energy for host–guest complexation can be obtained directly.

In general, host–guest complexation is a reversible process and it can be expressed through the following equation:

$$mH + nG \rightleftharpoons G_nH_m$$

where, m and n are the stoichiometry coefficients ($m, n = 1, 2, 3$, etc.). The interaction between the NBA and CB7 is of 1:2 nature obtained from ITC measurement. Here data obtained from ITC measurement was fitted by using two sets of sites binding model (Fig. 6). The formation constants value are $K_{1:1} = 3.80 \times 10^6\ M^{-1}$, $K_{1:2} = 1.99 \times 10^4\ M^{-2}$ and thermodynamic parameters determined by ITC are as follows $\Delta H_1 = -0.64 \times 10^4\ cal \cdot mol^{-1}$, $\Delta H_2 = -7.5 \times 10^4\ cal \cdot mol^{-1}$, $T\Delta S_1 = 0.26 \times 10^4\ cal \cdot mol^{-1}$ and $T\Delta S_2 = -6.91 \times 10^4\ cal \cdot mol^{-1}$.

From ΔH_1 and $T\Delta S_1$ value, it is cleared that the first step is both enthalpically as well as entropically favourable. The negative value of ΔH_1 may be due to the charge–dipole interaction (between positively charged nitrogen atom and carbonyl portal end of CB7) in addition to the hydrophobic contribution (between NEt_2 part of NBA and hydrophobic cavity of CB7). The positive value of $T\Delta S_1$ (measures the spontaneity of the system at temperature, T) is due to the release of high energy water molecules from the hydrophobic cavity of macrocyclic host, CB7 [31]. The hydrophobic cavity of this macrocycle does not allow to orient the trapped water molecules in energetically favourable H-bonding network. The potential energy of release trapped water molecules from the cavities of CB7 has maximum value among CBn. Hence, the excretion of these high-energy, water molecules from the CB7 cavity contribute to entropy gain in the first step. Deep penetration of the dye inside the hydrophobic cavity causes large number of water molecules release from hydrophobic cavity and significant entropic gain in first step is observed. For the entropy contribution in first step, the binding constant value in first step is found to be higher order as compared to second step. In second step, the change in enthalpy (ΔH_2) is negative but the value of $T\Delta S_2$ negative. The contribution of change in enthalpy (ΔH_2) is due to hydrogen bonding interaction between amine group ($-NH_2$) and the carbonyl portal end of CB7 and the negative value of $T\Delta S_2$ indicates that the system spontaneity decrease on formation of 1:2 complex. Hence, change in enthalpy (ΔH_2) is the driving force for the step second as $[\Delta H_2 - T\Delta S_2] < 0$. The binding value of Gibb's free energy in first step and second step are found to be $-2.14\ kJ \cdot mol^{-1}$ and $-1.41\ kJ \cdot mol^{-1}$, respectively ($\Delta G = \Delta H - T\Delta S$). Excited state nature and strength of interaction between NBA and CB7 is obtained from fluorescence measurement and by using ITC measurement we obtained the ground state binding nature and strength. From fluorescence and absorbance measurement, we observed that 1:1 complex is formed between NBA and CB7, whereas form ITC and ^1H NMR measurements it is observed that 1:2 complex is formed between NBA and CB7. The binding constant value obtained from spectroscopic and ITC measurement is different due to the fact that spectroscopy measured only local changes surrounding the dye molecule whereas, ITC measured a global change in the property [49]. The binding constant values may be the same if the hydration state of the interface is unchanged. Additionally, the binding constant values obtained from the fluorescence measurement and ITC measurements are similar if the host–guest complexation reaction process is passes through a two-state transition between free and bound dye molecules by following a lock and key or rigid body mechanism and the spectroscopic signal change must be reflected by the total population of free and bound molecules [50].

We have also studied the effect of addition of different surfactants and ionic liquid on the NBA–CB7 complex by using ITC measurement. It was observed that on gradual addition of 5.5 mM CTAB from the syringe to the aqueous solution of 0.05 mM NBA and 0.3 mM CB7 (inside the cell of the ITC instrument), initially kcal mol^{-1} of the injectant (Q) increases, it may be due to the dissociation of NBA–CB7 complex and

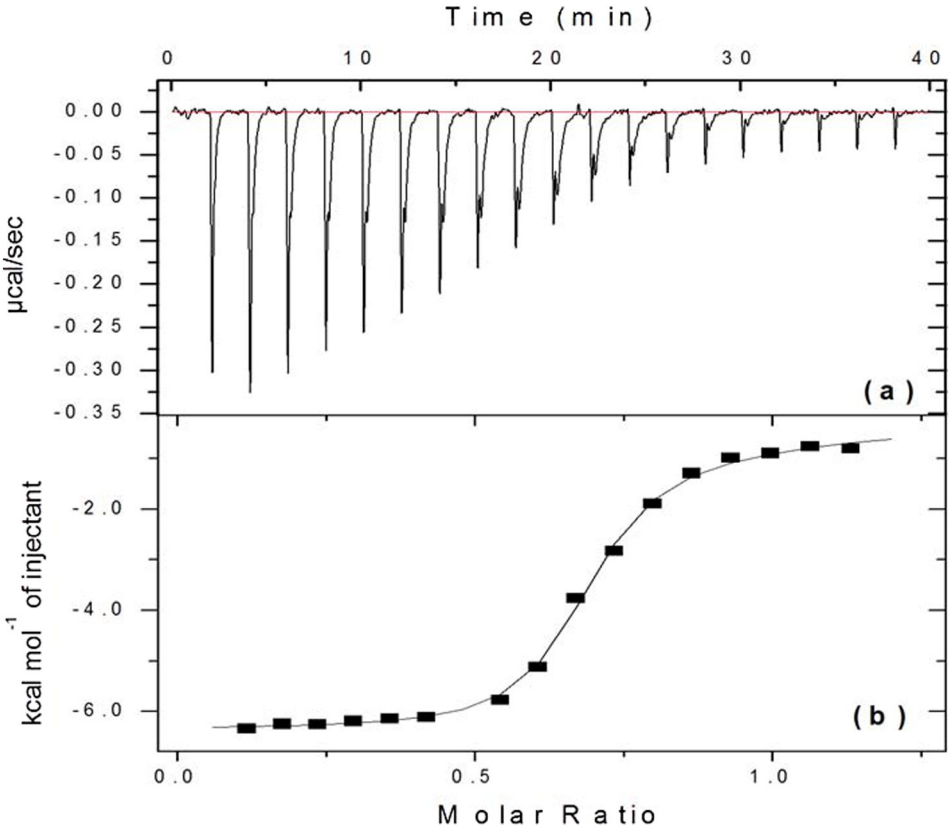


Fig. 6. (a) Heat flow versus time for the injection of 0.3 mM CB7 solution in 0.05 mM NBA solution at 298 K, (b) data points represent heat/mol versus CB7/NBA ratio and the solid line represents the line of best fit.

after reaching certain concentration of CTAB the *Q* value decreases, it may be due to the premiceller complex formed between surfactant and NBA (Fig. S7(a)). After completing this titration, further we added 0.01 M CTAB from syringe to the above solution (to reach the final concentration above *cmc*) and it was observed that the continuous decrease in kcal mol^{-1}. It indicates premicellar complex formation (CTAB < *cmc*) and CTAB micelle-NBA complex formation (CTAB > *cmc*) (Fig. S8(a)). All plots shown in Fig. S7 and Fig. S8 were obtained after blank subtraction.

On gradual addition of 0.056 M SDS to the aqueous solution of 2 mM CB7 and 0.05 mM NBA, initially the *Q* increases with increasing the value of mole fraction and after reaching some concentration *Q* value decrease and again after reaching certain concentration the value of *Q* increases (Fig. S7(b)). Initially the increase of *Q* value may be due to the dissociation of the complex and after that change in *Q* may be due to the contribution from the premiceller complex formation as well as electrostatic interaction of NBA with the monomer of SDS molecule. After completing this titration, we further added 0.1 M SDS to the final solution containing NBA, surfactant and CB7 to reach the concentration of SDS above cmc (Fig. S8(b)). It was observed that the *Q* value gradually increased due to the premicellar complex formation with NBA (SDS < *cmc*) as well as micelle-NBA complex formation (SDS > *cmc*). On addition of 0.22 M ionic liquid to the aqueous solution of NBA and CB7 initially the *Q* value increased and again further increased (Fig. S7(c)). Initially increase the value of *Q* can be ascribed to release of dye molecule from the hydrophobic core of CB7 and after that the change in *Q* is due to the formation of premicellar complex as well as electrostatic interaction of NBA with ionic liquid monomer. Further we added 0.75 M solution of ionic liquid through the syringe to the above solution containing NBA, CB7 and ionic liquid and observed that the value of kcal mol^{-1} of the injactant (Q) decreases with increasing molar ratio (IL/NBA), it may be due to the formation of premicellar complex (<*cmc*) and micelle–NBA complex (>*cmc*) (Fig. S8(c)).

3.9. Comparison of Stability of the Supramolecular Complexes in Ground and Excited States: Dissociation in Excited State or Effect of Radiation

The most dramatic feature of the host guest complex formation of the staining dye NBA with CB7 is that we observe 1:2 (guest:host) complex formation using the ^{1}H NMR and isothermal titration calorimetry (ITC) whereas, UV–Vis absorption spectra, steady state fluorescence emission spectra and time resolved emission spectra show 1:1 complex formation. This is really astonishing. While considering the complexation dynamics between a fluorescent host and a macrocycle, three factors must be considered. These are:

a. The rate of inclusion of the guest inside the cavity of macrocycle.
b. The rate of exclusion of the guest from the cavity of the macrocycle.
c. The rate of decay of the excited state of the probe.

When the excited state is short lived or in other words the excited state lifetime is short, the rate of excited state decay may be faster than either the rate of inclusion inside the host cavity or the rate of exclusion from the host cavity. Under such circumstances, only those guest molecules which will remain inside the hydrophobic cavity of macrocycle will absorb light to remain in excited state. Therefore, the rate of exclusion, rate of inclusion and the binding interaction of the guest molecule with the macrocycle will depend under such circumstances solely on the ground state binding interaction. However, electronic excitation of the guest by the absorption of light may also destabilize the complex and subsequently lead to the dissociation of the complex. This takes place when the excited state binding constant values are smaller than that of the ground state. For excited states with long decay time, it may be possible that the guests are excited outside the cavity of macrocycle to enter the cavity during the excited state lifetime. In such cases, the rates of inclusion and exclusion of the guest in the excited state can be significantly different from those of guest in the ground state. This accounts for the different value of the binding constant for the ground and excited states [11].

The determination of the binding constant provides a convenient way to determine extent of binding interaction between the dye and macrocycle. Determination of binding constant using fluorescence spectra provides the information regarding the binding interaction in the excited state whereas, the ground state binding interaction between the host and guest can be understood by determining the binding constant using ^{1}H NMR and isothermal titration calorimetry (ITC). A supramolecular host guest complex is extremely stable when the rate of inclusion (k_{in}) is significantly higher than the rate of exclusion (k_{out}). The binding constant value can be well represented as $K = \frac{k_{in}}{k_{out}}$ [51]. Similarly, this can be again represented as follows:

$$K = \frac{\tau_{out}}{\tau_{in}}$$

where τ_{out} is the time constant for the exclusion of the probe from the host cavity and τ_{in} stands for the time constant of the guest inclusion inside the cavity of the host. Comparison of ground state complex formation of the guest with the macrocycle using the ITC and absorption spectroscopy technique provides different information. UV–Vis spectral studies show that the host–guest complex is 1:1 in nature with binding constant value $5.8\ (\pm 5.1) \times 10^7\ M^{-1}$, the overall binding constant value ($K_{1:1}K_{1:2}$) determined using the ITC is 7.56×10^{10} with a 1:2 type binding. This shows that interaction of supramolecular guest: host complex with UV–Vis radiation causes the breakdown of 1:2 complex formed in the ground state itself. Here, we can make a rough estimate of the time spent by the dye molecule inside the cavity of CB7 as absence of any isobestic point in absorption spectra makes it impossible for us to determine the kinetics of complexation as described by Zhang et al. [52]. We have observed that on going from ground state to the excited state the binding constant ($6.2\ (\pm 1.9) \times 10^5\ M^{-1}$) decreases from that of ground state binding constant determined from both UV–Vis and ITC techniques. The time scale of the ^{1}H NMR spectra to observe the changes in δ values vary from microsecond to millisecond range. The very strong binding constant values determined from both UV–vis and ITC technique show that τ_{in} value is extremely small. Hence, the rate of inclusion of the dye inside the cavity of the CB7 is extremely high. The detection of presence of host guest complex in the ground state using ^{1}H NMR study is possible because the τ_{out} is almost comparable or higher than the time scale of ^{1}H NMR. The significant difference is observed in case of binding constant values in ground state and excited state. This is mainly due to the fact that the probe NBA excited outside the hydrophobic cavity of CB7 enters inside the cavity during the excited state lifetime thereby causing the difference of rate of inclusion and exclusion in the ground state and excited state.

Now another aspect of the difference of ground state and excited state complex formation is the difference of stoichiometry in ground and excited state. It can be presumed that while consider the detection of complex formation with ^{1}H NMR and ITC techniques, use of radiation is not required. While for absorption and steady state emission spectral studies, the initially formed 1:2 (guest:host) complex breaks down due to absorption of light. Thus we can detect only one kind of complex of 1:1 stoichiometry (guest: host) using absorption spectroscopy technique and this complex can be detected from the fluorescence study. The probable structure of the complex based on our study in shown in Scheme 2.

4. Conclusions

The supramolecular host–guest complexation of a red emitting dye, NBA with highly water soluble macrocycle CB7 and the surfactants and ionic liquid induced release of NBA from the hydrophobic cavity of CB7 was reported by using different spectroscopic and isothermal titration

calorimetric technique. On addition of CB7 to the aqueous solution of NBA prominent changes were observed in absorbance, fluorescence, fluorescence decay times and the changes were also observed on addition of surfactants and ionic liquid due to release of guest molecule from hydrophobic cavity of CB7. We have found that on addition of CB7 to the aqueous solution of NBA caused red shift in absorbance spectra along with increasing absorbance. At the same time addition of CB7 caused huge increase of the fluorescence intensity and the fluorescence quantum yield. It is demonstrated the formation of the complex between NBA and CB7. From Job's plot, it is confirmed that 1:1 complex is formed and the binding constant values were founded to be 5.8 $(\pm 5.1) \times 10^7\ M^{-1}$ from absorbance data and 6.2 $(\pm 1.9) \times 10^5\ M^{-1}$ from fluorescence data. From ^{1}H NMR studies, protons shift was observed in both the aliphatic and aromatic region in the presence of CB7. It indicates that the complex is formed between dye and CB7. Due to the formation of complex the fluorescence life time as well as the rotational correlation time increased. From ITC measurement it was observed that the complexation process is exothermic in nature and the process is both entalpically and entropically favourable. We have studied the release of NBA from the hydrophobic cavity of CB7 by using CTAB, SDS, TX-100 surfactants and [BMIM][OctS] ionic liquid. It is observed that neutral surfactant TX-100 has lesser effect on the host guest complex as compared to the ionic surfactants and the ionic liquid. In case of SDS and [BMIM][OctS] below *cmc* ion pair formation takes place between NBA and surfactant monomer as dye and surfactant/ionic liquid having opposite charges. Whereas, in the case of CTAB and TX-100 ion pair is not formed between dye and surfactants as CTAB and TX-100 are cationic and neutral surfactants, respectively. When the concentration reached to *cmc* then the complexation takes place between NBA and micelles formed by respective surfactants/ionic liquid.

Acknowledgements

All the authors are thankful to Indian Institute of Technology Patna (IIT Patna), India for the research facilities. S.A.A. and B.M. are thankful to IIT Patna for research fellowships. A.C. is thankful to CSIR, New Delhi (09/1023(0002)-EMR-I) for research fellowships.

Appendix A. Supplementary data

Supplementary data to this article can be found online at http://dx.doi.org/10.1016/j.jphotobiol.2016.05.009.

References

[1] K.I. Assaf, W.M. Nau, Cucurbiturils: from synthesis to high-affinity binding and catalysis, Chem. Soc. Rev. 44 (2015) 394–418.
[2] J. Lagona, B.D. Wagner, L. Isaacs, Molecular-recognition properties of a water-soluble cucurbit[6]uril analogue, J. Organomet. Chem. 71 (2006) 1181–1190.
[3] Y. Chen, Y. Liu, Cyclodextrin-based bioactive supramolecular assemblies, Chem. Sociol. Rev. 39 (2010) 495–505.
[4] N.J. Wheate, A.I. Day, R.J. Blanch, A.P. Arnold, C. Cullinane, J.G. Collins, Multi-nuclear platinum complexes encapsulated in cucurbit[n]uril as an approach to reduce toxicity in cancer treatment, Chem. Commun. (2004) 1424–1425.
[5] N. Basilio, L. García-Río, J.A. Moreira, M. Pessêgo, Supramolecular catalysis by cucurbit[7]uril and cyclodextrins: similarity and differences, J. Organomet. Chem. 75 (2010) 848–855.
[6] S. Gupta, R. Choudhury, D. Krois, U. Brinker, V. Ramamurthy, Cucurbituril adamantanediazirine complexes and consequential carbene chemistry, J. Organomet. Chem. 77 (2012) 5155–5160.
[7] J. Mohanty, H. Pal, A.K. Ray, S. Kumar, W.M. Nau, Supramolecular dye laser with cucurbit[7]uril in water, ChemPhysChem 8 (2007) 54–56.
[8] M.V.S.N. Maddipatla, L.S. Kaanumalle, A. Natarajan, M. Pattabiraman, V. Ramamurthy, Preorientation of olefins toward a single photodimer: cucurbituril-mediated Photodimerization of protonated Azastilbenes in water, Langmuir 23 (2007) 7545–7554.
[9] Y. Chen, Y. Liu, Cyclodextrin-based bioactive supramolecular assemblies, Chem. Soc. Rev. 39 (2010) 495–505.
[10] K.D. Daze, T. Pinter, C.S. Beshara, A. Ibraheem, S.A. Minaker, M.C.F. Ma, R.J.M. Courtemanche, R.E. Campbell, F. Hof, Supramolecular hosts that recognize methyllysines and disrupt the interaction between a modified histone tail and its epigenetic reader protein, Chem. Sci. 3 (2012) 2695–2699.
[11] B.D. Wagner, Hydrogen bonding of excited states in supramolecular host–guest inclusion complexes, Phys. Chem. Chem. Phys. 14 (2012) 8825–8835.
[12] K. Kim, Mechanically interlocked molecules incorporating cucurbituril and their supramolecular assemblies, Chem. Soc. Rev. 31 (2002) 96–107.
[13] A. Corma, H. García, P.M. Navajas, A. Primo, J.J. Calvino, S. Trasobares, Gold nanoparticles in organic capsules: a supramolecular assembly of gold nanoparticles and cucurbituril, Chem. Eur. J. 13 (2007) 6359.
[14] J. Kim, I.S. Jung, S.Y. Kim, E. Lee, J.K. Kang, S. Sakamoto, K. Yamaguchi, K. Kim, New cucurbituril homologues: syntheses, isolation, characterization, and X-ray crystal structures of cucurbit[n]uril (n) 5, 7, and 8), J. Am. Chem. Soc. 122 (2000) 540–541.
[15] G. Singh, M. Espiritu, Y. Shen, J.G. Hanlon, A.J. Rainbow, In vitro induction of PDT resistance in HT29, HT1376 and SK-N-MC cells by various photosensitizers, Photochem. Photobiol. 73 (2001) 651–656.
[16] C.W. Lin, J.R. Shulok, Y.K. Wong, C.F. Schanbacher, L. Cincotta, J.W. Foley, Photosensitization, uptake, and retention of phenoxazine Nile blue derivatives in human bladder carcinoma cells, Cancer Res. 51 (1991) 1109–1116.
[17] J. Morgan, W.R. Potter, A.R. Oseroff, Comparison of photodynamic targets in a carcinoma cell line and its mitochondrial DNA-deficient derivative, Photochem. Photobiol. 71 (2000) 747–757.
[18] M. Wainwright, H. Mohr, W.H. Walker, Phenothiazinium derivatives for pathogen inactivation in blood products, J. Photochem. Photobiol. B 86 (2007) 45–58.
[19] A.A. Gorodetsky, W.J. Hammond, M.G. Hill, K. Slowinski, J.K. Barton, Scanning electrochemical microscopy of DNA monolayers modified with Nile blue, Langmuir 24 (2008) 14282–14288.
[20] Y.I. Yang, H.Y. Hong, I.S. Lee, D.G. Bai, G.S. Yoo, J.K. Choi, Detection of DNA using a visible dye, Nile blue, in electrophoresed gels, Anal. Biochem. 280 (2000) 322–324.
[21] Q.Y. Chen, D.H. Li, Y. Zhao, H.H. Yang, Q.Z. Zhua, J.G. Xu, Interaction of a novel red-region fluorescent probe, Nile blue, with DNA and its application to nucleic acids assay, Analyst 124 (1999) 901–906.
[22] A. Grofcsik, M. Kubinyi, W.J. Jones, Intermolecular photoinduced proton transfer in Nile blue and oxazine 720, Chem. Phys. Lett. 250 (1996) 261–265.
[23] R.K. Mitra, S.S. Sinha, S.K. Pal, Interactions of Nile blue with micelles, reverse micelles and a genomic DNA, J. Fluoresc. 18 (2008) 423–432.
[24] K. Hirakawa, K. Ota, J. Hirayama, S. Oikawa, S. Kawanishi, Nile blue can photosensitize DNA damage through electron transfer, Chem. Res. Toxicol. 27 (2014) 649–655.
[25] B.R. Raju, S. Naik, P.J.G. Coutinho, M.S.T. Gonçalves, Novel Nile blue derivatives as fluorescent probes for DNA, Dyes Pigments 99 (2013) 220–227.
[26] G.B. Dutt, S. Doraiswamy, N. Periasamy, B. Venkataraman, Rotational reorientation dynamics of polar dye molecular probes by picosecond laser spectroscopic technique, J. Chem. Phys. 93 (1990) 8498–8513.
[27] S.A. Ahmed, A. Chatterjee, B. Maity, D. Seth, Supramolecular interaction of a cancer cell photosensitizer in the nanocavity of cucurbit[7]uril: a spectroscopic and calorimetric study, Int. J. Pharm. 492 (2015) 103.
[28] M. Sayed, M. Sundararajan, J. Mohanty, A.C. Bhasikuttan, H. Pal, Photophysical and quantum chemical studies on the interactions of oxazine-1 dye with cucurbituril macrocycles, J. Phys. Chem. B 119 (2015) 3046–3057.
[29] Y. Liu, B. Han, Y. Chen, Inclusion complexation of acridine red dye by calixarenesulfonates and cyclodextrins: opposite fluorescent behavior, J. Organomet. Chem. 65 (2000) 6227–6230.
[30] J. Mohanty, A.C. Bhasikuttan, W.M. Nau, H. Pal, Host–guest complexation of neutral red with macrocyclic host molecules: contrasting pKa shifts and binding affinities for cucurbit[7]uril and β-cyclodextrin, J. Phys. Chem. B 110 (2006) 5132–5138.
[31] F. Biedermann, V.D. Uzunova, O.A. Scherman, W.M. Nau, A.D. Simone, Release of high-energy water as an essential driving force for the high-affinity binding of cucurbit[n]urils, J. Am. Chem. Soc. 134 (2012) 15318–15323.
[32] D. Banik, J. Kuchlyan, A. Roy, N. Kundu, N. Sarkar, Stimuli-sensitive breathing of cucurbit[7]uril cavity: monitoring through the environment responsive fluorescence of 1′-Hydroxy-2′-acetonaphthone (HAN), J. Phys. Chem. B 119 (2015) 2310–2322.
[33] J. Mohanty, N. Thakur, S.D. Choudhury, N. Barooah, H. Pal, A.C. Bhasikuttan, Recognition-mediated light-up of thiazole orange with cucurbit[8]uril: exchange and release by chemical stimuli, J. Phys. Chem. B 116 (2012) 130–135.
[34] M. Pessêgo, J.P.D. Silva, J.A. Moreira, L. García-Río, Differences in cucurbit[7]uril: surfactant complexation promoted by the cationic head group, ChemPlusChem 78 (2013) 1058–1064.
[35] S.D. Choudhury, N. Barooah, V.K. Aswal, A.C. Bhasikuttan, H. Pal, J. Mohanty, Stimuli-responsive supramolecular micellar assemblies of cetylpyridinium chloride with cucurbit[5/7]urils, Soft Matter 10 (2014) 3485–3493.
[36] K. Das, B. Jain, H.S. Patel, Nile blue in Triton-X 100/benzene–hexane reverse micelles: a fluorescence spectroscopic study, Spectrochim. Acta A 60 (2004) 2059–2064.
[37] P. Pal, H. Zeng, G. Durocher, D. Girard, R. Giasson, L. Blanchard, L. Gaboury, L. Villeneuve, Spectroscopic and photophysical properties of some new rhodamine derivatives in cationic, anionic and neutral micelles, J. Photochem. Photobiol. A Chem. 98 (1996) 65–72.
[38] M. Sarkar, S. Poddar, Studies on the interaction of surfactants with cationic dye by absorption spectroscopy, J. Colloid Interface Sci. 221 (2000) 181–185.
[39] D. Seth, S. Sarkar, N. Sarkar, Dynamics of solvent and rotational relaxation of Coumarin 153 in a room temperature ionic liquid 1-butyl-3-methylimidazolium octyl sulfate forming micellar structure, Langmuir 24 (2008) 7085–7791.
[40] Z. Miskolczy, K. Sebok-Nagy, L. Biczok, S. Gökturk, Aggregation and micelle formation of ionic liquids in aqueous solution, Chem. Phys. Lett. 400 (2004) 296–300.
[41] A. Douhal, Photophysics of Nile blue A in proton-accepting and electron-donating solvents, J. Phys. Chem. 98 (1994) 13131–13137.

[42] T. Kobayashi, Y. Takagi, H. Kandori, K. Kemnitz, K. Yoshihara, Femtosecond intermolecular electron transfer in diffusionless, weakly polar systems: Nile blue in aniline and N,N-dimethylaniline, Chem. Phys. Lett. 180 (1991) 416–422.
[43] P. Thordarson, R.G.E. Coumans, J.A.A.W. Elemans, P.J. Thomassen, J. Visser, A.E. Rowan, R.J.M. Nolte, Allosterically driven multicomponent assembly, Angew. Chem. Int. Ed. 43 (2004) 4755–4759.
[44] S. Nigam, G. Durocher, Spectral and photophysical studies of inclusion complexes of some neutral 3H-indoles and their cations and anions with β-cyclodextrin, J. Phys. Chem. 100 (1996) 7135–7142.
[45] D. Maity, T. Govindaraju, Pyrrolidine constrained bipyridyl-dansyl click luoroionophore as selective Al^{3+} sensor, Chem. Commun. 46 (2010) 4499–4501.
[46] B. Kalman, N. Clarke, L.B.A. Johansson, Dynamics of a new fluorescent probe, 2,5,8,11-tetra-*tert*-butylperylene, in solution, cubic lyotropic liquid crystals, and model membranes, J. Phys. Chem. 93 (1989) 4608–4615.
[47] P. Sen, D. Roy, S.K. Mondal, K. Sahu, S. Ghosh, K. Bhattacharyya, Fluorescence anisotropy decay and solvation dynamics in a nanocavity: Coumarin 153 in methyl β-cyclodextrins, J. Phys. Chem. A 109 (2005) 9716–9722.
[48] J. Edwards, Molecular volumes and the Stokes–Einstein equation, J. Chem. Educ. 47 (1970) 261–270.
[49] M.T. Rehman, H. Shamsi, A.U. Khan, Insight into the binding mechanism of imipenem to human serum albumin by spectroscopic and computational approaches, Mol. Pharm. 11 (2014) 1785–1797.
[50] A. Chatterjee, B. Maity, D. Seth, Supramolecular interaction between a hydrophilic coumarin dye with macrocyclic hosts: spectroscopic and calorimetric study, J. Phys. Chem. B 118 (2014) 9768–9781.
[51] B.D. Wagner, The use of coumarins as environmentally-sensitive fluorescent probes of heterogeneous inclusion systems, Molecules 14 (2009) 210–237.
[52] X. Zhang, G. Gramlich, X. Wang, W.M. Nau, A joint structural, kinetic, and thermodynamic investigation of substituent effects on host–guest complexation of bicyclic azoalkanes by β-cyclodextrin, J. Am. Chem. Soc. 124 (2002) 254–263.