J Incl Phenom Macrocycl Chem (2007) 59:183–190
DOI 10.1007/s10847-007-9313-4

ORIGINAL ARTICLE

Photophysical behaviours of some 2-styrylindolium dyes in aqueous solutions and in the presence of cyclodextrins

Asya Petinova · Stefan Metsov · Ivan Petkov · Stefan Stoyanov

Received: 20 October 2006 / Accepted: 13 March 2007 / Published online: 25 April 2007

Abstract Host–guest inclusion type association between native β-cyclodextrin and randomly substituted methyl-β-CD and two 2-styrylindolium cationic dyes, e.g. 1,3,3-trimethyl-2-(4-diethylaminostyryl)-3H-indolium iodide (D1) and 1,3,3-trimethyl-2-[4-(N-2-cyanoethyl,N-methyl)-aminostyryl]-3H-indolium iodide (D2), are reported. The described indolium derivatives belong to the rarely investigated class of unsymmetrical polymethines. The complex formation was studied in aqueous buffer solutions with two pH values (7.2 and 3) by means of absorption and steady-state fluorescence spectroscopy. The association equilibrium constant (K), the molar absorptivity and the stoichiometry of the complexes were evaluated using the modified Benesi-Hildebrand approach. The complex stability was affected by the pH of the solution and by the type of CD. The results obtained indicate that D1 forms 1:1 complexes with both β-CD and Me-O-β-CD, whereas D2 does not form stable complexes with Me-O-β-CD and in acidic medium. The fluorescent intensity of D1 in the presence of CDs increases over four times relative to the intensity of the pure dye solutions, presumably via inclusion of the dye into the cyclodextrin cavity due to rigidity of the structure.

A. Petinova (✉) · I. Petkov
Department of Organic Chemistry, Faculty of Chemistry, Sofia University, James Bourchier Blvd. 1, Sofia 1164, Bulgaria
e-mail: asya_petinova@chem.uni-sofia.bg

S. Metsov · S. Stoyanov
Department of Organic Chemical Technology, Faculty of Chemistry, Sofia University, James Bourchier Blvd. 1, Sofia 1164, Bulgaria

Keywords Cyclodextrin · Inclusion complexes · Optical spectroscopy · Stability constant · Styrylindolium dyes

Introduction

Cyclodextrins (CDs) are cyclic oligosaccharides produced from starch. They are cone-shaped capsules with an internal hydrophobic cavity and hydrophilic exterior. The size of the cavity depends on the number of glucose units and the most common CDs are those with 6, 7 and 8 units (5–8 Å), named α-, β- and γ-CD, respectively. This feature accounts for their unique ability to form inclusion or host–guest complexes with a large variety of guest molecules in aqueous medium. Only physical forces work in the complexes, with no covalent bonding [1]. The stability of inclusion complexes in aqueous solutions is mainly due to van der Waals forces and hydrophobic interactions in which the solvent water pushes the hydrophobic side of the guest molecule into the hydrophobic CD cavity [2]. Therefore, methylation of the hydroxyl groups of CDs would lead to a change in the stability, solubility, and structure of the inclusion complexes.

The complexation phenomenon often involves remarkable variations in the photophysical and photochemical properties of the guest molecule because of the environmental difference between the CD interior and the outer environment. The key observation is that the formation of supramolecular complexes of analytes with CDs provokes an increase of their fluorescence quantum yield [3, 4] or even the appearance of room-temperature phosphorescence [5, 6]. Also, the rates of photochemical reactions decrease notably when CD inclusion complexes are formed due to the geometric restriction imposed on the guest molecule

[3, 7]. Besides, the binding of guest molecules within the host cyclodextrin is not fixed or permanent but is rather a dynamic equilibrium. The size, shape and polarity of the guest molecule relative to the CD low-polar inner cavity are critical parameters for the complexation and for the stability of host–guest complexes.

The styryl dyes with electron donor–acceptor moieties on either side of the styryl fragment are particularly attractive for their spectral sensitivity towards the local environment and for their optical and electronic properties. They are widely used in medicine for the fluorescent labelling of biomolecules [8, 9], as dyes for optical records for image and information storage, as active materials in dye lasers, in trace metal detection, etc. Relatively few works have been devoted to the spectral study of CD complexes with hemicyanine dyes, and these focus mainly on their photostability. Kasatani [10, 11] and Matsuzawa [12] observed enhancement of dimerization and an increase of the light stability of cyanine dyes with a long polymethine chain in the presence of β- and γ-CD. Styrylindolium and styrylpyridinium cationic dyes are hemicyanine dyes which have been used as chromophores by modification of crown ethers [13, 14] and calixarenes [15] for spectrophotometric detection of metal ions and surfactants. The most important photochemical process for these hemicyanine dyes is the reversible geometrical E-Z isomerization. It proceeds from the first exited singlet state and competes with the photophysical process fluorescence.

The purpose of the present work is to investigate the effect of the environment on the spectral properties of two styrylindolium dyes (Scheme 1) in aqueous buffer solutions (pH 7.20 and pH 3) using absorption and steady-state fluorescence spectroscopies. The reason to choose these dye molecules as spectral probe is the fact that they are assumed to be very sensitive to environmental changes which will enable us to investigate their inclusion complexation behaviour with CDs. We chose native β-CD and its derivative methyl-β-CD (randomly substituted) as hosts in order to evaluate the influence that substitution might have on complex stability and the stoichiometric ratio of the inclusion complexes. Besides, the dyes are chosen so as to be structurally similar because we wanted to study whether the substituents in the guest molecule affect the inclusion complex formation. Dialkylamino and cyano groups are well known and are used as donor and acceptor groups, respectively. Therefore, we chose D1 so as to have a strongly electron-donating group ($-N(Et)_2$) and D2, where one of the ethyl substituents is modified with a strongly electron-withdrawing group (–CN). Our interest was focused on the inclusion of dye molecules into the CD cavity since these species could be candidates for optical sensors for organic molecules. We hope that such a systematic study would help understand, predict and control the chemical and physical properties of analogous hemicyanine dyes included in CDs.

It should be pointed out that, in the concentration range investigated, there is no evidence for aggregation of the dyes. A possible reason for this could be steric hindrance occurring at the methyl groups in the heterocyclic ring.

Experimental

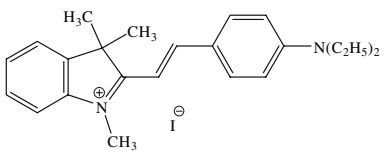
Reagents

All experiments were performed with analytical grade chemicals. β-CD (obtained from Fluka, purum ≥ 99%, HPLC) and Me-O-β-CD (average M = 1310 g/mol) purchased from Aldrich were used as received. The dyes D1 and D2 were synthesized as described earlier and their structures were proved by means of IR and ^{1}H NMR spectroscopies [16]. The purity of the dyes in these samples had been established by thin layer chromatography and optical spectroscopy. Distilled water and phosphate buffer solution (pH 7.20) and citrate-hydrochloric acidic buffer (pH 3) obtained from Merck were used to control the pH-value.

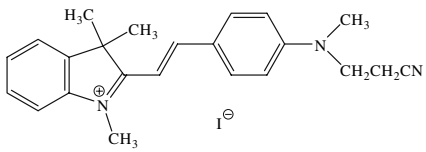
Apparatus

All absorption measurements were made with a Thermo Spectronic instrument Unicam UV 500 UV-Visible double-beam spectrophotometer using 1 cm quartz cells. Data processing was performed on a Hewlett Packard computer with Vision-Pro software. Samples were scanned from 190 to 900 nm. The fluorescence measurements were performed with Cary Eclipse Fluorescence spectrophotometer from Varian. The excitation source was a xenon flash lamp.

Scheme 1 Structures of D1 and D2



1,3,3-trimethyl-2-(4-diethylaminostyryl)-3H-indolium iodide (D1)



1,3,3-trimethyl-2-[4-(N-2-cyanoethyl,N-methyl)-aminostyryl]-3H-indolium iodide (D2)

Excitation and emission bandwidths were both set at 5 nm, and the excitation wavelengths were 548 and 514 nm for D1 and D2 respectively. All experiments were carried out at room temperature.

Solution preparation

We used phosphate buffer aqueous solution to make the sample solutions. Concentrated stock solutions (2.2×10^{-4} M) were prepared separately for each dye by dissolving the required amount of the compound in water. β-CD and Me-O-β-CD standard solutions with concentration 3.54×10^{-3} M and 3.51×10^{-3} M respectively were prepared. Working solutions or solutions with lower concentration were obtained by appropriate dilutions of these standard solutions. In order to study the influence of the matrix on the dye intensity in this medium, several cyclodextrin solutions were prepared, by maintaining a constant concentration of dyes (2.2×10^{-5} M for D1 and 2.1×10^{-5} M for D2) and varying the CD concentration. An aliquot of the guest stock solution was placed in a 10 mL volumetric flask, the appropriate amount of cyclodextrin solution was added to give the final CD concentration and it was diluted with the solvent to the final volume of 10.0 mL. These measurements were carried out after at least 5-hour dark storage.

Results and discussion

Absorption spectra

The absorption spectra were generated by exposing dye solutions to light at various wavelengths. The presence of both donor and acceptor ends in the parent chromophore system (dialkylamino- and 3H-indolium quaternary nitrogen atoms respectively) is responsible for the appearance of a long-wave absorption charge-transfer band (CT) (Fig. 1). Electronic charge is transferred from the amino-substituent of the phenyl ring to the nitrogen atom of the indolium ring upon excitation to the lowest excited state. D1 was found to exhibit a broad band with λ_{max} value at 548 nm and D2 at 514 nm. These bands can be assigned to π–π^* transition involving the whole electronic system of the compounds with considerable CT character. When the CD concentration was increased, the intensity of the peaks decreased. The spectral decrease is accompanied by a hypsochromic shift of the CT band relative to the spectrum of the two dyes alone (538 nm and 510 nm respectively), which may be due to the transportation of the dye molecules from the polar solvent (water) into the relatively apolar cavity of cyclodextrin to form an inclusion complex. Furthermore, the absorption spectra exhibit well defined isosbestic points at about 530 nm for D1 and 485 nm for D2. The presence of isosbestic points suggests the existence of various absorbing species in the solutions and formation of host–guest complexes with stoichiometry 1:1. In general, D1 is causing stronger shift than D2.

In ground state and in neutral medium, these unsymmetrical styryl dyes might be represented by two extreme resonance structures [17] (Fig. 2). In contrast to symmetrical cyanines, the two resonance structures *A* and B are not equivalent. This suggests some bond alternation leading to a relatively greater half-bandwidth of the longest-wavelength absorption band.

According to Brooker [18] and Suzuki [19], the two extreme mesomeric structures (Fig. 2) of styryl dyes are not energetically equivalent. Structure *A* predominates in the ground state, whereas the contribution of structure B is greater in the first excited singlet state.

The increase of the acidity of the medium resulted in considerable colour change in dye D1 due to the protonation of the dye molecules at the atom with the highest electron density. This is the nitrogen atom of the amino-substituent. In this case intramolecular CT does not occur and the long-wavelength band disappears. A new absorption band appeared in the UV region due to localized π–π^* at the cost of the CT band (Fig. 3). It is most likely that the charge transfer was inhibited due to protonation of the amino-nitrogen atom; thus the free electron pair could not

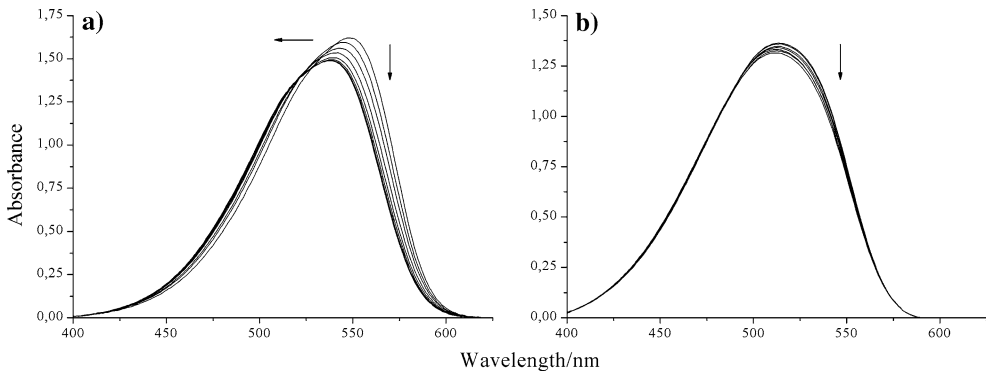
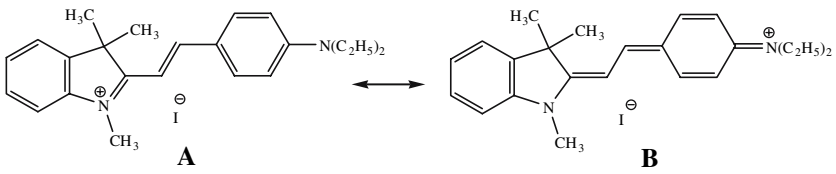


Fig. 1 Absorption spectra of (**a**) D1 (2.2×10^{-5} M) and (**b**) D2 (2.1×10^{-5} M) in water (pH 7.2) containing various concentrations of Me-O-β-CD

Fig. 2 Extreme resonance structures (A and B) of D1



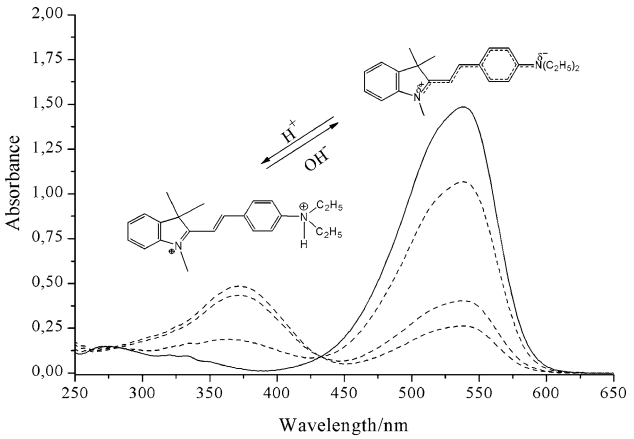


Fig. 3 Absorption spectrum of D1 aqueous solution upon addition of 5% HCl

take part in delocalization with another unsaturated part of the molecule and the dye D1 absorbed in a shorter-wavelength region.

The intensity of the long-wavelength band at 548 nm decreases, whereas a new band at around 375 nm appears. A clear isosbestic point is observed in the absorption spectrum, and that suggests that there are only two absorbing species under these conditions which turn into each other.

D2 contains an electron-donating substituent an amino group, too, but instead of having two alkyl substituents, it is connected to an electron-withdrawing functional group (–CN) which attracts the lone electron pair on the N-atom and disturbs the conjugation. Consequently, D2 absorbs at considerably shorter wavelength (514 nm) than the structurally similar dye D1, and the acidic medium does not affect the absorption spectrum of the compound. The spectral characteristics of the dyes in aqueous solution in the presence of various amounts of β-CD are listed in Table 1. The acidity of the medium does not influence the position of the absorption and emission maximum for both dyes.

We studied complex formation in aqueous buffer solution with pH 3 by adding increasing amounts of β-CD and Me-O-b-CD molecules. As we saw (Fig. 3), there were two absorbing systems of the D1 dye in this medium non-protonated (548 nm) and protonated (375 nm), which were in equilibrium. However, the presence of a host molecule influences this equilibrium. The increase of host concentration leads to a decrease of short-wavelength intensity and to an increase of long-wavelength absorbance (Fig. 4). There is a well defined isosbestic point at about 426 nm, which indicates the conversion of the protonated into non-protonated form. Moreover, the increase in the intensity of the conjugated form is accompanied by a hypsochromic

Table 1 Absorption and fluorescence maxima of D1 (2.2×10^{-5} M) and D2 (2.1×10^{-5} M) at various β-CD concentrations and various pH values

Dye	[β-CD] mol L^{-1}	pH 7.2			pH 3		
		$\lambda(abs)_{max}$ (nm)	Log ε_0	$\lambda(em)_{max}$ (nm)	$\lambda(abs)_{max}$ (nm)	Log ε_0	$\lambda(em)_{max}$ (nm)
D1	0	548	4.90	596	548	4.69	593
					367	4.08	
	0.00018	543	4.88	593	545	4.72	592
					367	4.05	
	0.00071	539	4.87	588	541	4.76	589
					364	3.91	
	0.0018	538	4.86	586	538	4.80	586
					362	3.79	
D2	0	514	4.79	581	514	4.81	581
	0.00018	514	4.79	581	514	4.81	581
	0.0007	512	4.78	580	513	4.80	581
	0.0018	510	4.78	579	512	4.79	581

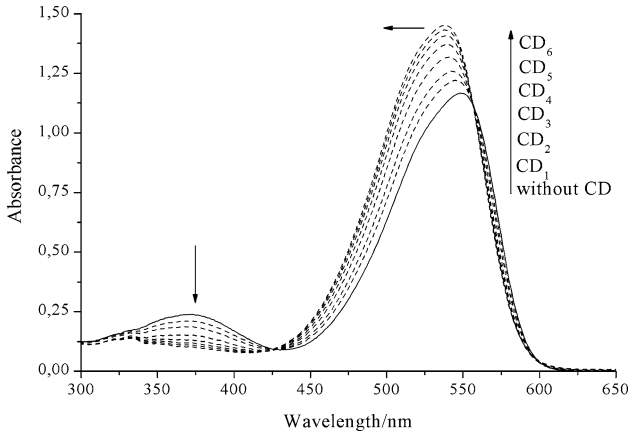


Fig. 4 Absorption spectra of D1 (2.2×10^{-5} M) in aqueous buffer solution (pH 3) at various β-CD concentrations: $[CD_1] = 1.76 \times 10^{-4}$; $[CD_2] = 3.52 \times 10^{-4}$; $[CD_3] = 7.04 \times 10^{-4}$; $[CD_4] = 1.06 \times 10^{-3}$; $[CD_5] = 1.41 \times 10^{-3}$; $[CD_6] = 1.76 \times 10^{-3}$; $[CD_7] = 2.11 \times 10^{-3}$ M

shift of the maximum at about 538 nm. As we mentioned above, this corresponds to inclusion complex formation between D1 and CD molecules. The existence of an isosbestic point at about 556 nm is consistent only with one type of interaction with stoichiometry 1:1.

Such behaviour in acidic medium shows that CDs interact with the D1 molecule and furthermore, this interaction is only with the non-protonated form. By adding host molecules, an inclusion complex between this form and the macrocycles is formed and the equilibrium moves to the left (Fig. 5).

Based on these spectral observations on the complex formation of D1, we assumed that the dye penetrates the cyclodextrin cavity by means of its phenyl ring together with the N,N-diethylamino substituent and not with the heterocycle, and that the former group remains hidden for the hydrogen ions of the bulk solution.

The inclusion complexation with CDs is a result of the simultaneous contribution of a wide variety of weak interactions, of which van der Waals, hydrophobic interactions and hydrogen bond formation are the most important ones. Thus it is easy to explain why the inclusion of charged molecules (or a part of them) in the cavity is less favoured than that of uncharged molecules.

The participation of the phenyl ring into complexation was confirmed also by the absorption spectra of D2 with various hosts. At pH 7.2, and with the increase of CD concentration, D2 undergoes fewer changes in the intensity and the shift of the maximum compared to D1. In view of the fact that the absorption spectra were measured under the same conditions and that the two dyes differ from one another only in the substituent of the phenyl ring, it is evident that it is precisely this cycle that takes part in complex formation.

Determination of binding constants

The most popular method for estimating the stiochiometry, the equilibrium constant (K) and the molar absorptivity of non-covalent complexes on the basis of spectral changes is the Benesi-Hildebrand method [20]. Since the CDs are spectroscopically inert, we used spectroscopically active guests which were undergo changes in the absorption spectrum when the host concentration is varied. In fact, the dye molecules and the inclusion complexes coexisted in the solution, therefore, UV-vis absorption, as shown on Fig. 1, with various CD concentrations, is superposition of the absorptions of dye monomers and D–$(CD)_n$ inclusion complexes, where n is the number of CDs in the inclusion complex. If we assume that the ratio of inclusion complexes ($n = 1$) is 1:1, the system can be described as in eq. 1:

$$D + CD \overset{K_{11}}{\leftrightarrows} D - CD \qquad K_{11} = \frac{[D - CD]}{[D][CD]} \tag{1}$$

where D and D–CD denote the dye molecule and its complex respectively. In the presence of excess CD, the equilibrium concentration of CD approximates the initial

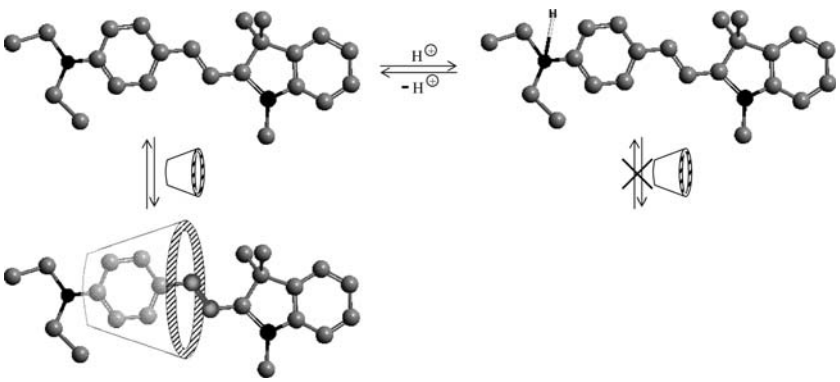


Fig. 5 Disturbance of the equilibrium after addition of CD and the proposed structure of the 1:1 inclusion complex

concentration of CD $[CD]_0$. The spectral data in Fig 1 can be analysed according to the Benesi-Hildebrand equation (eq. 2):

$$\frac{[D]_0[CD]_0}{\Delta A} = \frac{1}{K_{11}\Delta\varepsilon_{11}} + \frac{[CD]_0}{\Delta\varepsilon_{11}} \quad (2)$$

Here $[D]_0$ is the total concentration of the dyes, $\Delta\varepsilon_{11}$ is the difference in the molar absorptivity between the free and the complexed dye molecules ($\Delta\varepsilon_{11} = \varepsilon_0 - \varepsilon_{11}$) and ΔA is the change in the absorption intensity of the dye solutions when CD is added. A linear relationship between $[D]_0[CD]_0/\Delta A$ and $[CD]_0$ will be obtained if the stoichiometry of the complex is 1:1. A well defined straight line with good correlation coefficients confirmed that the assumption of a 1:1 ratio of D1 and D2 and CDs is correct (Fig. 6). From the slope and intercept ratio of the plot we obtained the corresponding association constants with the various pH values and these are shown in Table 2.

The data reported in Table 1 show that the native β-CD at pH 7.2 seems to be a better host than methyl-β-CD because of the change in the size and shape of the macrocycle. Apparently, the methylation of hydroxyl groups increases the water solubility of host molecules by distorting the hydrogen bonds on the CD rim, but, on the other hand, it narrows the inner diameter of the cavity and reduces the stability of the complexes [21, 22]. Besides, the substitution of the OH-groups in Me-O-β-CD reduces the possibility of hydrogen bonding with the amino-N-atom of the dyes, which additionally lowers the complex stability. These features are most probably the reason for the substantially reduced stability constants of both D1 and D2 with β-CD and Me-O-β-CD. In contrast to the case of D1, D2 seems not to form inclusion association with the hosts, except for D2-β-CD complexes at pH 7.2. The changes in the absorption and fluorescence spectra are insignificant and no reasonable fit to eq. 2 was obtained. Furthermore,

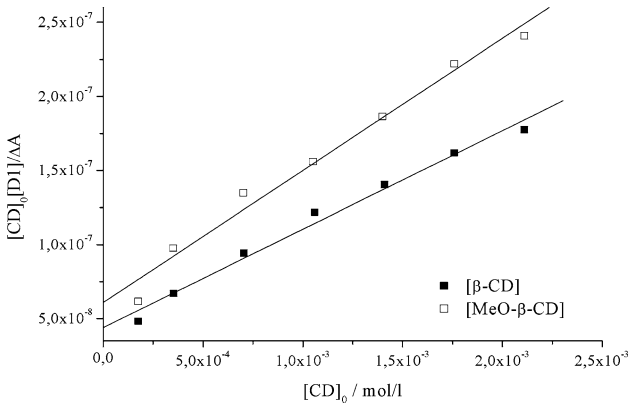


Fig. 6 Determination of the association constant of D1 with β-CD and Me-O-β-CD in aqueous solution (pH 3)

Table 2 Stability constants K_{11} (mol^{-1} L) for 1:1 complexes of dye molecules and CDs at pH 7.2 and pH 3

Dye	CD	K_{11} (pH 7.2)	K_{11} (pH 3)
D1	β-CD	2167	1693
	Me-O-β-CD	1354	1350
D2	β-CD	760	–
	Me-O-β-CD	–	–

the considerably low K_{11} values of D2 with β-CD compared to D1 with β-CD are due not so much to steric hindrances as to the electrostatic repulsion between the basic inner surface of the cavity and the cyano group in the amino-substituent. Moreover, since the CN group reduces the basicity of the amino-N-atom, the host–guest complexes cannot be stabilized through hydrogen bonding with the primary hydroxyls of CDs.

With both β-CD and Me-O-β-CD, the stability constants of the inclusion complexes of the dyes were reduced in acidic media. Since protonated D1 cannot interact with the CD cavity and it interacts only with the diethylamino-group, D1 gives relatively low binding ability with β-CD at pH 3 (K_{11} = 1693 M^{-1}).

Fluorescence measurement

It is well known that polar solvents result in a drastic reduction of the fluorescence quantum yield and an increase of the photoisomerization quantum yield. The ratios of the fluorescence intensities of complexed and free dye molecules in aqueous buffer solutions are presented in Table 3. In our case, D1 shows relatively low fluorescence in aqueous solution,which increases drastically with the addition of host molecules, whereas the fluorescence intensity of D2 does not vary substantially in the presence of CD (Fig. 7). Table 1 depicts the emission maxima of D1 and D2 in neutral and in acidic medium with various β-CD concentrations. The fluorescence spectra of the two dyes are identical at pH 7.2 and pH 3 – the position of the emission maximums remain unaffected and there are changes only in the intensity.

Upon addition of CD, the fluorescence intensity of D1 is enhanced, which is accompanied by a shift of the fluorescence maximum to shorter wavelengths at about 10 nm. This finding indicates that the dye molecule changes its environment and passes into a less polar medium. The change in the fluorescence intensity is most likely caused by the decrease of the competitive photochemical and radiationless rate constants of D1 in the complex. When the molecule penetrates the CD cavity, its structure becomes more rigid and the dye loses the absorbed energy mainly through emission of light. Thus the fluorescence intensity

Table 3 Ratios of the fluorescence intensities of included and free dye molecules at various pH values*

Dye	I_f^{CD}/I_f^0			
	β-CD (1.77 x 10^{-3} M)	Me-O-β-CD (1.75 x 10^{-3} M)	β-CD (1.76 x 10^{-3} M)	Me-O-β-CD (1.76 x 10^{-3} M)
D1 (2.2 x 10^{-5} M)	4.8[a]	4.5[a]	3.3[b]	3.2[b]
D2 (2.1 x 10^{-5} M)	1.3[a]	1.1[a]	1.1[b]	1.1[b]

*Aqueous buffer solutions with pH 7.2 and pH 3: I_f^0-fluorescence intensity of the dyes alone; I_f^{CD}-fluorescence intensity of included dyes; [a] I_f^{CD}/I_f^0 at pH 7.2; [b] I_f^{CD}/I_f^0 at pH 3

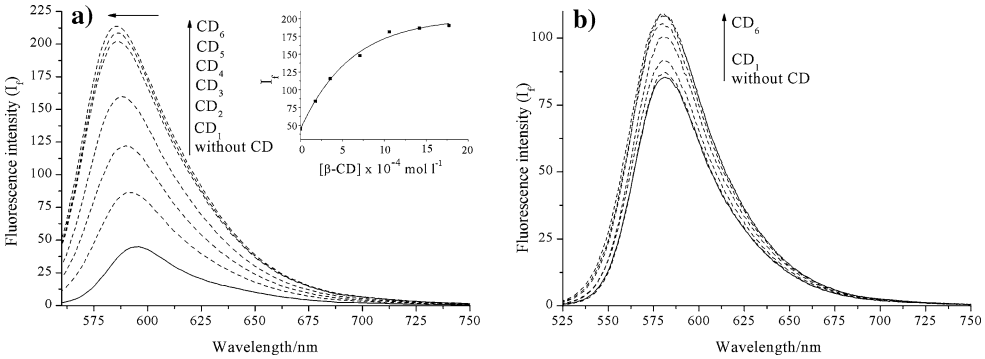


Fig. 7 Fluorescence intensity of D1 (**a**) (2.2×10^{-5} M) and D2 (**b**) (2.1×10^{-5} M) at various β-CD concentrations (pH 7.2): (—) without β-CD, (- - -) with β-CD: [β-CD_1] = 1.77×10^{-4}, [β-CD_2] = 3.54×10^{-4}, [β-CD_3] = 7.08×10^{-4}, [CD_4] = 1.06×10^{-3}, [β-CD_5] = 1.42×10^{-3}, [β-CD_6] = 1.77×10^{-3} M; Inset a): the plot of fluorescence intensity of D1 vs. β-CD concentration

of D1 increases over four times compared to the intensity of D1 alone (Table 3). Further enhancement of CD concentration does not lead to any appreciable changes, which could be related to the achievement of an optimum interaction dye cyclodextrin; an equilibrium is reached, which is not influenced by the change of host concentration (Fig. 7 a).

The addition of host to D2 solution does not provoke significant changes in fluorescence intensity. There is a blue shift of the maximum only at about 1–2 nm, and 1.3 times higher intensity in the presence of β-CD and solution pH of 7.2 (Fig. 7 b). Fluorescence spectra in the presence of β-CD (pH 3) and Me-O-β-CD (both pH 7.2 and pH 3) do not show any shift of the maximum and the intensity does not change in a regular manner but fluctuates around identical values. This evidence shows that D2 forms much weaker complexes with β-CD compared to D1 and does not form complexes with modified β-CD. This assumption is in agreement with the data from the absorption spectra.

Conclusion

We used the absorption and fluorescence spectroscopies to study host–guest complexes between native and modified CD and two styrylindolium dyes in neutral and in acidic medium. The decrease in the absorption of the dyes along with the hypsochromic shift and the isosbestic points observed in the presence of varying CD concentrations provide supporting evidence for the formation of CD/dye complexes. The shifts indicate that a less polar environment gradually replaces the aqueous medium surrounding the dye, as the cyclodextrin host is added to the solution. We determined which parts of the dye molecules interact with the two hosts. The stoichiometry of all complexes, obtained by a Benesi-Hildebrand plot, was 1:1, and the binding constants were determined using the changes of absorption intensity with various CD concentrations. The K_{11} values for Me-O-β-CD were remarkably lower than those for β-CD, which is related mainly to steric hindrance due to the reduction of cavity diameter by methylation and to the lowering of the ability for hydrogen bonding between primary hydroxyls and amino-N-atoms. The inclusion complexes were also characterized by an enhancement of the fluorescence signal (over four times for D1) relative to the free dye in aqueous buffer solution due to the fact that the emission quenching intramolecular motions were very restricted. The fluorescence intensity of D1 increases upon interaction with β-CD and Me-O-β-CD and the formation of inclusion complexes, whereas D2 forms unstable complexes with β-CD and even does not interact with modified β-CD.

Acknowledgement Financial support from the National Science Foundation—Bulgaria is gratefully acknowledged.

References

1. Connors, K.A.: The stability of cyclodextrin complexes in solution. Chem. Rev. **97**, 1325–1357 (1997).
2. Liu, L., Guo, Q.X.: The driving forces in the inclusion complexation of cyclodextrins. J. Incl. Phenom. Mac. Chem. **42**, 1–14 (2002).
3. Coly, A., Aaron, J.J.: Cyclodextrin-enhanced fluorescence and photochemically-induced fluorescence determination of five aromatic pesticides in water. Anal. Chim. Acta **360**, 129–141 (1998).
4. Maafi, M., Laassis, B., Aaron, J.J., Mahedero, M.C., Munoz de la Pena, A., Salinas, F.: Photochemically induced fluorescence investigation of a β-cyclodextrin: Azure A inclusion complex and determination of analytical parameters. J. Incl. Phenom. Mol. Recogn. Chem. **22**, 235–247 (1995).
5. Gonzalez, M.A., Lopez, M.H.: Determination of fluorene in sea-water by room temperature phosphorescence in organised media. Analyst **123**, 2217–2221 (1998).
6. Scypinski, S., Cline Love, L.J.: Room-temperature phosphorescence of polynuclear aromatic hydrocarbons in cyclodextrins. Anal. Chem. **56**, 322–327 (1984).
7. Duveneck, G.L., Sitzmann, E.V., Eisenthal, K.B., Turro, N.J.: Picosecond laser studies on photochemical reactions in restricted environments: the photoisomerization of trans-stfibene complexed to cyciodextrins. J. Phys. Chem. **93**, 7166 (1989).
8. Patonay, G., Salon, J., Sowell, J., Strekowski, L.: Noncovalent labeling of biomolecules with red and near-infrared dyes. Molecules **9**, 40–49 (2004).
9. Deligeorgiev, T.G., Zaneva, D.A., Kim, S.H., Sabnis, R.W.: Preparation of monomethine cyanine dyes for nucleic acid detection. Dyes and Pigments **37**, 205–211 (1998).
10. Ohashi, M., Kasatani, K., Shinohara, H., Sato, H.: Molecular mechanics studies on inclusion compounds of cyanine dye monomers and dimers in cyclodextrin cavities. J. Am. Chem. Soc. **112**, 5824–5830 (1990).
11. Kasatani, K., Ohashi, M., Kawazaki, M., Sato, H.: Cyanine dye-cyclodextrin systems. Enhanced dimerization of the dye. Chem. Lett. 1633–1636 (1987).
12. Matsuzawa, Y., Tamura, S., Matsuzawa, N., Ata, M.: Light stability of a β-cyclodextrin inclusion complex of a cyanine dye. J. Chem. Soc. Faraday Trans. **90**(23), 3517–3520 (1994).
13. Mitewa, M., Mateeva, N., Antonov, L.: Spectrophotometric investigation on the complexation between chromo- and flouroionophores containing aza-15crown-5 moiety and alkaline and alkaline-earth metal ions. Quim Analitica **16**, 153–162 (1997).
14. Gromov, S.P., Fedorova, O.A., Alfimov, M.V., Druzhinin, S.I., Rusalov, M.V., Uzhinov, B.M.: Crown-containing styryl dyes. 14. Synthesis, luminescence, and complexation of the trans-isomers of chromogenic 15-crown-5-ethers. Izv. Akad. Nauk., Ser. Khim. **10**, 2003–2008 (1995).
15. Nishida, M., Ishii, D., Yoshida, I., Shinkai, S.: Molecular association of water-soluble calixarenes with several stilbene dyes and its application to the facile determination of cationic surfactant concentrations. Bull. Chem. Soc. Jpn. **70**, 2131–2140 (1997).
16. Metsov, S., Dudev, T., Koleva, V.: Infrared and NMR study of some 2-styrylindolium dyes. J. Mol. Str. **350**, 241–246 (1995).
17. Kiprianov, Al., Ushenko, I.K.: Colour of the organic dyes and its plane geometry. Zh. Obshch. Khim. **20**, 514, (1950).
18. Brooker, L.G.S.: Absorption and resonance in dyes. Rev. Mod. Phys. **14**, 275–293 (1942).
19. Suzuki, H.: Electronic Absorption Spectra, Geometry of Organic Molecules. Academic Press, New York (1967), pp. 367.
20. Benesi, H.A., Hildebrand, J.H.: A spectrophotometric investigation of the interaction of iodine with aromatic hydrocarbons. J. Am. Chem. Soc. **71**, 2703–2707 (1949).
21. Yoshida, N., Fujita, Y.: Dynamic aspects in host-guest interactions. 3. Kinetics and mechanism for molecular recognition by hexakis(2,6-di-o-methyl)-α-cyclodextrin of some azo guest molecules. J. Phys. Chem. **99**, 3671–3677 (1995).
22. Sueishi, Y., Hishikawa, H.: Complexation of 4-dimethylaminoazobenzene with various kinds of cyclodextrins: effects of cyclodextrins on the thermal cis-to-trans isomerization. Int. J. Chem. Kin. **34**, 481–487 (2002).