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Solvent effects in the interaction of methyl-β-cyclodextrin with solvatochromic merocyanine dyes

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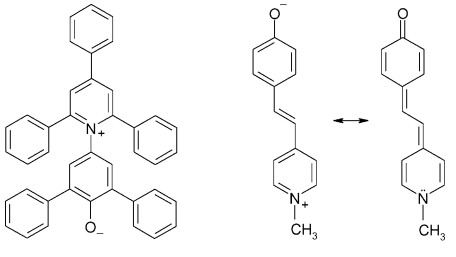
The UV-vis spectroscopic behavior of dyes: 2,6-diphenyl-4-(2,4,6-triphenylpyridinium-1-yl)phenolate (**1**) and 4-[(1-methyl-4(1*H*)-pyridinylidene)-ethylidene]-2,5-cyclohexadien-1-one (**2**) was investigated in solutions of methyl-β-cyclodextrin (methyl-β-CyD), using water, methanol, ethanol, propan-2-ol, butan-1-ol, acetone, acetonitrile, *N*,*N*-dimethylformamide (DMF), dimethyl sulfoxide (DMSO), *N*,*N*-dimethylacetamide (DMA), chloroform and dichloromethane as solvents. In aqueous solutions of dye (**2**) the addition of M-β-CD leads to a bathochromic shift (of the maximum absorption), showing that the probe was transferred to a microenvironment of lower polarity and suggesting the formation of a 1 : 1 dye (**2**) : CyD inclusion complex, with a binding constant of 128.5 ± 3.5 $dm^3\ mol^{-1}$. Data for dye **2** in alcohols showed hypsochromic shifts, which increased in the following order: methanol < ethanol < propan-2-ol < butan-1-ol. These observations appear to reflect dye–solvent interactions through hydrogen bonding. If dye–solvent interactions are strong, the CyD–dye interactions are consequently weak, but the latter increase in importance when the dye–solvent interaction becomes weaker. With hydrogen-bond accepting solvents, data for both dyes showed clearly increasing hypsochromic shifts following the order: DMSO < DMA < DMF < acetone < acetonitrile. This order is exactly the inverse of the increasing order of basicity of the medium. This indicates that the dominant factor for the observed effects in these solvents is the solvent–CyD interaction through hydrogen bonding involving the hydroxyl groups of the CyD and the basic groups of the solvents. These interactions diminish in intensity if the basic character of the medium is reduced, increasing the capability of the dye to interact with the CyD using its phenoxide donor moiety. The largest hypsochromic shifts were obtained in chloroform (66.0 nm) and dichloromethane (67.5 nm) with dye **1** after addition of methyl-β-CyD. In these specific situations, solvents display weak basic and acid properties, that enhanced CyD–dye interactions to such an extent that association complexes formed through hydrogen bonding could be detected (K_{11} values of 24.8 ± 4.9 $dm^3\ mol^{-1}$ in dichloromethane and 66.1 ± 8.0 $dm^3\ mol^{-1}$ in chloroform).

Introduction

Cyclodextrins (CyDs) or cycloamyloses are cyclic oligosaccharides comprised of 6 or more glucose units connected by α-1,4 bonds. These cyclic molecules are versatile receptors for a variety of organic and inorganic compounds,[1,2] since they have hydrophobic cavities with a hydrophilic exterior, with the diameter of the cavities (4.7–8.3 Å) being determined by the number of glucose units. The most studied cyclodextrins are α-, β- and γ-CyD with 6, 7 and 8 glucose units, respectively, which are commercially available in their natural or modified form. The ability of CyDs to form inclusion complexes in solution and in the solid state has led to a great variety of research, due not only to their potential industrial applications,[3] but also to developments in bioorganic chemistry, in the study of enzyme action[4] and in the comprehension of the biological receptor–substrate interactions.[4–6] CyDs are also applied in chromatography[7] and capillary electrophoresis.[8] Since the number of potential applications involving CyDs is very large and one of the main factors responsible for the formation of inclusion complexes is related to their hydrophobic cavity, studies on their micropolarity are fundamental for the comprehension of complex formation and solvent effects related to these processes.

Physicochemical investigations of medium polarity are commonly performed by means of chemical probes.[9–12] An important example is the dye 2,6-diphenyl-4-(2,4,6-triphenylpyridinium-1-yl)phenolate (**1**), which is one of the most commonly employed solvatochromic dyes in studies in solution.[9–11] Another well-known solvatochromic dye is 4-[(1-methyl-4(1*H*)-pyridinylidene)-ethylidene]-2,5-cyclohexadien-1-one (**2**), better known as Brooker's merocyanine.[11,13] These dyes have been used in recent years in several studies related to solvatochromism,[9–11,13] halochromism,[14] mixed solvents[15,16] and microheterogeneity in solution.[17]

Despite the large number of studies on the behavior of different classes of dyes in solution in the presence of CyDs,[18,19] there are no reports in the literature regarding studies with solvatochromic dyes, such as with the well-known probes **1** and **2**. These studies are important in order to make estimates about the micropolarity of the cavity of the CyDs.[2] In this paper we describe the investigation of the UV-vis spectroscopic behavior of dyes **1** and **2** in solutions containing methyl-β-CyD. Solutions of the dyes with methyl-β-CyD in hydrogen-bond donating (HBD) solvents (water, methanol, ethanol, propan-2-ol and butan-1-ol), hydrogen-bond accepting (HBA) solvents (acetone, acetonitrile, *N*,*N*-dimethylformamide (DMF), dimethyl sulfoxide (DMSO) and *N*,*N*-dimethylacetamide (DMA)) and chlorinated solvents (chloroform and dichloromethane) were investigated and the data obtained are explained using a model based on microscopic interactions involving the dye, the CyD and the solvent.



1 **2**

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Table 1 'Polarity' parameters of pure solvents and the variation in molar transition energy for dyes **1** and **2** in the absence and in the presence of methyl-β-CyD

Solvent	E_T^N[a]	β[b]	α[b]	DN[c]	ΔE_T(**1**)[d]	ΔE_T(**2**)[d]
Water	1.00	0.47[e]	1.17	138[f]	−26.4	−12.8
Methanol	0.762	0.62	0.93	126	0.00	0.00
Ethanol	0.654	0.77	0.83	134	0.00	0.795
Butan-1-ol	0.586	0.88	0.79	—	2.65	5.65
Propan-2-ol	0.546	0.95	0.76	88.3	3.68	1.21
DMSO	0.444	0.76	0.00	125	1.59	0.879
DMF	0.386	0.69	0.00	111	3.31	4.06
DMA	0.377	0.76	0.00	116	2.05	2.68
Acetone	0.355	0.48	0.08	71.1	10.8	13.3
Acetonitrile	0.460	0.31	0.19	59.0	8.83	18.4
Chloroform	0.259	0.00	0.44	16.7	16.2	[g]
Dichloromethane	0.309	0.00	0.30	6.69	18.1	[g]

[a] Ref. 11. [b] Ref. 20. [c] Ref. 22 and 23, values given in kJ mol^{-1}. [d] This study, ΔE_T(dye) = E_T(dye)$_{CyD}$-E_T(dye), E_T(dye) and E_T(dye)$_{CyD}$ being values obtained in the absence and in the presence of 0.02 mol dm^{-3} methyl-β-CyD, respectively, in kJ mol^{-1} at 25 °C. [e] For bulk water, ref. 21. [f] For bulk water. [g] The dye is insoluble.

Results and discussion

Table 1 displays the Reichardt normalized E_T^N values,[11] Kamlet–Taft α (solvent hydrogen-bond donor acidity) and β (solvent hydrogen-bond acceptor basicity) parameters,[20,21] that are commonly used to describe solvent polarity and its hydrogen bonding properties. Table 1 also gives the Guttmann donor number (DN) values,[22,23] that represent an attempt to measure solute/electron pair donor–solvent interactions (the larger the DN value, the greater the ability of the solvent to donate an electron pair or, in other words, the greater its donicity). The solvents used in this study are classified as three groups. The first group is composed of the HBD solvents, with high E_T^N and α values, that decrease in magnitude with an increase in the alkylic chain. It has been argued that the pyridinium phenolate **1** used in the construction of the E_T^N scale can interact specifically with the solvent through hydrogen bonding, and these values can reflect the HBD capacity of the medium.[24] At the same time, for these solvents, an increase in β and DN values can be observed with an increase in the alkylic chain of the solvent. The second group embraces HBA solvents, which are very good hydrogen bond acceptors, displaying high β and DN values (except for acetonitrile), and poor HBD solvents, with low α and E_T(30) values. The latter group comprises the chlorinated solvents chloroform and dichloromethane, which are both poor HBA and HBD solvents.

Table 1 also shows the spectral behavior of the solutions of dyes in many solvents with the addition of methyl-β-CyD, expressed in the form of ΔE_T(**1**) and ΔE_T(**2**) values for dyes **1** and **2**, respectively. They were calculated from the expression ΔE_T(dye) = E_T(dye)$_{CyD}$ − E_T(dye), E_T(dye) and E_T(dye)$_{CyD}$ being the molar transition energies for the dye given in kJ mol^{-1} in the absence and in the presence of methyl-β-CyD, respectively. In the case of the ΔE_T data in Table 1, the concentration of CyD is 0.02 mol dm^{-3}. The data clearly show that a bathochromic shift occurs in the aqueous solutions of the dyes after the addition of methyl-β-CyD, as expressed by the negative ΔE_T values. A spectral shift was not observed for solutions of **1** and **2** in methanol and **1** in ethanol, while a very small hypsochromic shift (1.7 nm) was observed for the solutions of **2** in ethanol after addition of the CyD. Positive ΔE_T values (hypsochromic shifts) were obtained for all other solutions of the dyes with the addition of the CyD. Values of ΔE_T for dye **2** could not be obtained in chloroform and in dichloromethane because this dye is not soluble in these solvents.

Karl Fischer titrations revealed the presence of 10% (wt) water in the CyD. Since hypsochromic shifts in the solvatochromic bands of dyes **1** and **2** are obtained if water is added to organic cosolvents, due to its specific interaction with the phenolic moiety of the dye, it could be possible that the hypsochromic shifts observed were caused by water molecules present in the CyD. Therefore, it is important to compare the data from this study with others reporting the effect of 10% (wt) water added to solutions of the dyes in the solvents. Data are available from the literature for dyes **1**[25] and **2**[16] in many aqueous mixtures and some of these data were used in the calculation of the shifts that occur with the addition of 10% water in alcohols (methanol, ethanol and propan-2-ol) and HBA solvents (DMSO, DMF, acetone and acetonitrile). These data are compiled in Table 2 and they show that for these dyes, although the addition of water in general causes a hypsochromic shift of the solvatochromic bands, it is clear that a part of the shift is due to the interaction between the CyD and the dye. Acetone and acetonitrile, in particular, gave the greatest shifts due to the presence of the CyD. For instance, a hypsochromic shift of 30 nm is obtained for the solutions of **1** in acetone in the presence of the CyD when the effect due to the presence of water is discarded. The nature of these interactions is discussed in the following sections.

Interaction of the dyes with methyl-β-CyD in water

The large bathochromic shifts observed for aqueous solutions containing methyl-β-CyD 0.02 mol dm^{-3} correspond to 50.3 and 22.2 nm for dyes **1** and **2**, respectively. It has long been known that a hypsochromic shift of the solvatochromic band of **1** and **2** occurs with increasing solvent polarity, due to the greater stability of the ground state of the structure in relation to its excited state.[9–11] Thus, data obtained in water show that the probes are transferred to a low polarity microenvironment with the addition of the CyD, and suggest that inclusion of the dyes in the hydrophobic cavity of the CyD occurs. The larger bathochromic shift obtained for dye **1** can be explained by the fact that this dye displays a more accentuated solvatochromism than dye **2** and also because dye **1** is more hydrophobic than **2**, interacting more efficiently with the hydrophobic cavity of the CyD.

Table 2 Comparison between the λ_{max} values for the solvatochromic bands of dyes **1** and **2** in the pure solvents, in the solvents with 10% (w/w) of water and in the solvents with 0.02 mol dm^{-3} methyl-β-CyD

	λ_{max} (dye **1**)/nm			λ_{max} (dye **2**)/nm		
Solvent	Pure solvent[a]	With 10% of water[b]	With methyl-β-CyD[c]	Pure solvent[d]	With 10% of water[d]	With methyl-β-CyD[c]
Methanol	516	514	516	485	485	485
Ethanol	551	548	551	514	513	512
Propan-2-ol	591	600	580	546	544	543
DMSO	634	635	629	568	568	566
DMF	662	653	650	583	579	572
Acetone	677	668	638	588	578	552
Acetonitrile	627	616	599	562	556	517

[a] From ref. 11. [b] Calculated from ref. 25. [c] This study. [d] From ref. 16.

Fig. 1 depicts the spectral behavior of dye **2** in aqueous solution with the addition of increasing amounts of methyl-β-CyD. It can be seen that the solvatochromic band of **2** at 445.7 nm suffers a large bathochromic shift and a reduction in absorbance after the addition of the CyD: with a CyD concentration of 0.02 mol dm^{-3}, the band shifts to 467.9 nm. Fig. 1 also shows an isosbestic point at 461.0 nm, suggesting the presence of a 1 : 1 dye : CyD inclusion complex. Absorbances at 443.6 nm were collected from the spectra and are given in Fig. 2, which shows that more than 95% of the dye is included at a CyD concentration of 0.04 mol dm^{-3}. The inclusion constant was calculated from these data, firstly using eqn. (3), that gave a K_{11} value of 128.2 dm^3 mol^{-1} ($r^2 = 0.9996$ and sd $= 1.58 \times 10^{-3}$). Experimental data were then fitted with a nonlinear regression to eqn. (1), providing a very good fit and giving a K_{11} value of 128.5 ± 3.5 dm^3 mol^{-1} ($\chi^2 = 4.12 \times 10^{-6}$). Similar experiments could not be carried out with dye **1** due to its very low solubility in pure water.

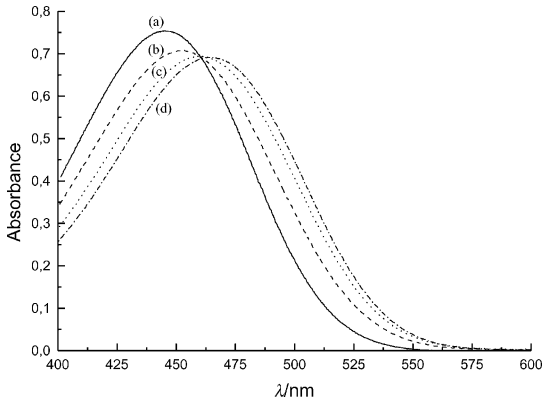

Fig. 1 UV-vis spectra at 25 °C of aqueous solutions of dye **2** (4.7×10^{-5} mol dm^{-3}) containing methyl-β-CyD in concentrations of (a) zero, (b) 3.0×10^{-3}, (c) 8.0×10^{-3} and (d) 1.4×10^{-2} mol dm^{-3}.

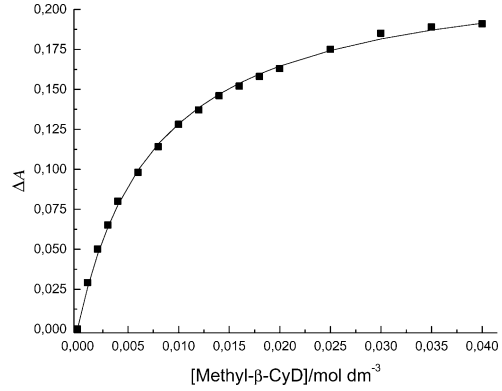

Fig. 2 Variation in the absorbance of the solvatochromic band of dye **2** in water with the addition of increasing amounts of methyl-β-CyD. The concentration of **2** was 4.7×10^{-5} mol dm^{-3} and absorbances were collected at 443.6 nm. (—) Curve fitted with eqn. (1).

Solvent effects on the behavior of dyes 1 and 2 in alcohols and in HBA solvents in the presence of methyl-β-CyD

Data from Table 1 allow a comparative study of the solutions of dyes **1** and **2** in the absence and in the presence of methyl-β-CyD in alcoholic solutions and also in solutions of HBA solvents. Data for alcohols show that the ΔE_T(**2**) values follow the increasing order: methanol < ethanol < propan-2-ol < butan-1-ol. This order is exactly the decreasing order for the $E_T{}^N$ values and the Kamlet–Taft a values, suggesting that the acidity of the medium is the key factor in the understanding of the behavior of the dyes in alcoholic solutions. Thus, the behavior of dye **2** in methanol, with no shift observed with the addition of the CyD, can be explained by the fact that the solvent interacts so strongly with the dye and with the CyD through hydrogen bonding that dye–CyD interactions cannot occur. With the decreasing acidic character of the medium, together with the increase in the alkylic chain of the alcoholic component, the interaction of the solvent with the phenolic oxygen of the dye and with the hydroxyl groups in the CyD diminishes and the CyD–dye interactions through hydrogen bonding can be evidenced.

Figs. 3 and 4 illustrate the effect of the addition of increasing amounts of methyl-β-CyD on the solvatochromic bands of dyes **1** and **2** in HBA solvents. They show clearly the following increasing order in the hypsochromic shifts for both dyes: DMSO < DMA < DMF < acetone < acetonitrile. This order matches exactly the inverse increasing order in the β and DN values for these solvents, *i.e.*, the higher the basicity of the solvent the lower the observed ΔE_T value. Solvents with high HBA properties (high DN and β values) can interact strongly with HBD species. Since CyDs have hydroxyl groups in their structures, strong CyD–solvent interactions are to be expected. These interactions diminish in intensity if the basic character of the medium is reduced, increasing the capability of the dye to interact with the CyD using its phenoxide donor moiety. Thus, the strength of the CyD–dye interactions depends on the solvent and can be adjusted by the selection of the solvating medium.

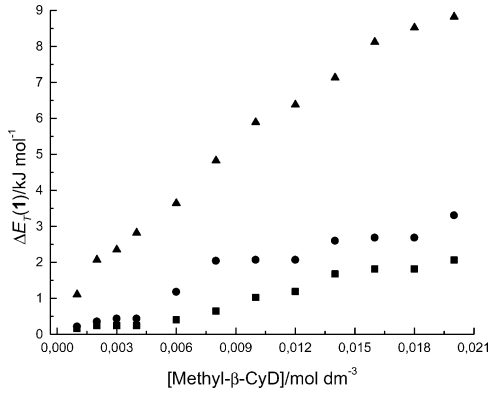

Fig. 3 Variations in the molar transition energies of the solvatochromic band of dye **1** in DMA (■), DMF (●) and acetonitrile (▲) solutions with the addition of increasing amounts of methyl-β-CyD at 25 °C. The concentration of **1** was 1.0×10^{-4} mol dm^{-3}.

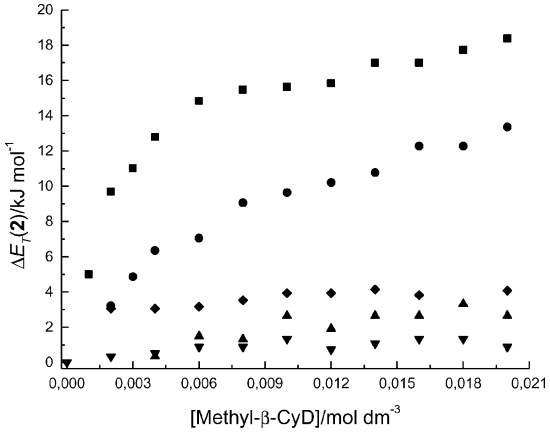

Fig. 4 Variations in the molar transition energies of the solvatochromic band of dye **2** in DMSO (▼), DMA (▲), DMF (◆), acetone (●) and acetonitrile (■) solutions with the addition of increasing amounts of methyl-β-CyD at 25 °C. The concentration of **2** was 4.7×10^{-5} mol dm^{-3}.

A comparison between the ΔE_T values in Table 1 for dyes **1** and **2** shows that the largest hypsochromic shifts generally occur with dye **2**. In principle, this could be due to a difference in

the basicity of the probes, with the most basic probe interacting more strongly with the CyD and displaying a more pronounced spectral shift. Data from the literature corroborate partially this explanation, since the pK_a values for protonated dyes **1** and **2** are 8.4[26] and 8.6,[27] respectively. Coleman and Murray[28] showed that 2,6-di-*tert*-butyl-4-methylphenol is not capable of interacting with dye **1** through hydrogen bonding in acetonitrile due to the steric hindrance of the *tert*-butyl groups in the phenol and of the phenyl groups in the phenoxide moiety of the dye. Since these groups are absent in dye **2**, a more effective interaction with the CyD and more substantial hypsochromic shifts are expected.

Influence of the addition of methyl-β-CyD to solutions of dye 1 in chloroform and in dichloromethane

The experimental data relating to dye **1** (Table 1) show that the most significant hypsochromic shifts after addition of methyl-β-CyD were obtained with chloroform (66.0 nm) and dichloromethane (67.5 nm). The variations in the maximum of the solvatochromic band of **1** as a function of the CyD concentration in these solvents are depicted in Fig. 5. The low polarity of these chlorinated solvents makes them able to solvate the hydrophobic dye by non-specific interactions, but the ability of these solvents to interact, although weakly, with the phenolic group of **1** through hydrogen bonding has been reported,[29] since α values of these solvents cannot be neglected. On the other hand, dye **1** is able to interact strongly with hydroxylic solvents through hydrogen bonding, as confirmed by the high E_T^N values obtained in these solvents.[11] In addition, studies with **1** in mixed solvents show that this probe is preferentially solvated by the hydroxylic component in mixtures of alcohols with dichloromethane or chloroform,[25,30] which can be attributed to the specific dye–alcohol interaction through hydrogen bonding. Moreover, the zero β values of chloroform and dichloromethane rule out any specific interaction of the CyD with these solvents acting as hydrogen-bond acceptors. Since methyl-β-CyD has hydroxyl groups in its structure, it is reasonable to assume that these groups can interact specifically with the dye by means of hydrogen bonding, since the specific interaction of the chlorinated solvent with the phenolic group in the dye is weak. Therefore, the large shifts may be due to three combined effects: both weak solvent–CyD and solvent–dye interactions and strong CyD–dye interactions.

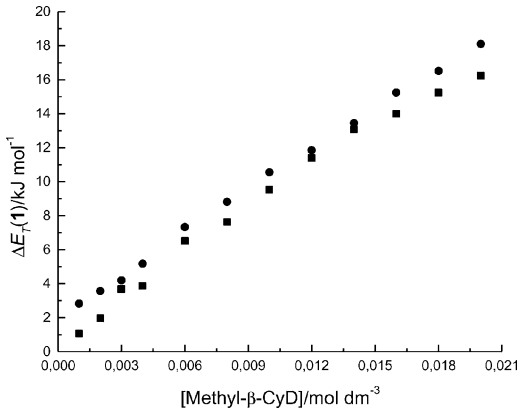

Fig. 5 Variations in the molar transition energies of the solvatochromic band of dye **1** in chloroform (■) and dichloromethane (●) solutions with the addition of increasing amounts of methyl-β-CyD at 25 °C. The concentration of **1** was 1.0×10^{-4} mol dm^{-3}.

Fig. 6 displays the spectral behavior of a solution of dye **1** in dichloromethane with the addition of increasing amounts of methyl-β-CyD. The solvatochromic band of **1** at 702.5 nm suffers a large hypsochromic shift after the addition of the CyD changing to 640.4 nm with a CyD concentration of 0.018 mol dm^{-3}. An isosbestic point occurs at 602.5 nm, suggesting the presence of a 1 : 1 dye : CyD complex formed through hydrogen bonding. Absorbance values obtained at 691.2 nm allowed an estimate of the association constant of the complex. The use of eqn. (2) gave a K_{11} value of 22.1 dm^3 mol^{-1} ($r^2 = 0.999$ and sd $= 6.7 \times 10^{-2}$). Experimental data were then fitted to eqn. (1), giving a K_{11} value of 24.8 ± 4.9 dm^3 mol^{-1} ($\chi^2 = 9.0 \times 10^{-5}$).

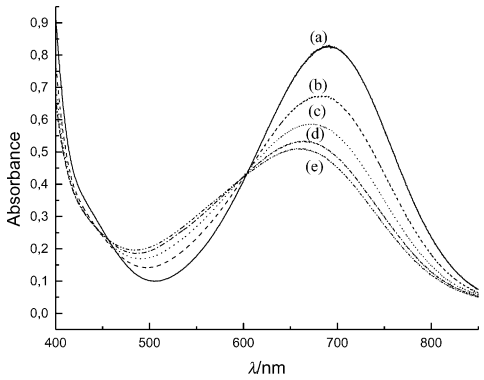

Fig. 6 UV-vis spectra at 25 °C of solutions of dye **1** (1.0×10^{-4} mol dm^{-3}) in dichloromethane containing methyl-β-CyD in concentrations of (a) zero, (b) 4.7×10^{-3}, (c) 8.4×10^{-3}, (d) 1.1×10^{-2} and (e) 1.2×10^{-2} mol dm^{-3}.

A similar situation was observed using chloroform as the solvent. In this case, an isosbestic point was observed at 630.5 nm and the absorbance values at 719.8 nm were used in the construction of Fig. 7, which shows the variation in the solvatochromic band of **1** with the addition of increasing amounts of methyl-β-CyD. The association constant was calculated from these data, firstly using eqn. (2), giving a K_{11} value of 61.2 dm^3 mol^{-1} ($r^2 = 0.998$ and sd $= 9.8 \times 10^{-2}$). A data fit with eqn. (1) gave a K_{11} value of 66.1 ± 8.0 dm^3 mol^{-1} ($\chi^2 = 5.0 \times 10^{-5}$).

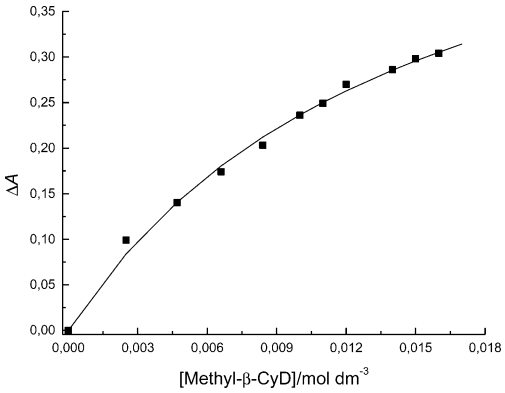

Fig. 7 Variations in the absorbance of the solvatochromic band of dye **1** in chloroform with the addition of increasing amounts of methyl-β-CyD. The concentration of **1** was 1.0×10^{-4} mol dm^{-3} and absorbances were collected at 719.8 nm. (——) Curve fitted with eqn. (1).

Conclusions

The UV-vis study of the solvatochromic bands of merocyanine solvatochromic dyes **1** and **2** after addition of methyl-β-CyD in different HBD and HBA solvents showed that solvatochromic dyes take part in strong specific interactions, such as hydrogen bonding and the hydrophobic effect. Spectral shifts for the dyes with the addition of methyl-β-CyD in the studied solvents show that, except for water, a model can be built in order to explain the data, which considers dye–solvent, CyD–solvent and CyD–dye interactions. According to this model, solvents with high α values interact strongly with the dye through hydrogen bonding, thus weakening the CyD–dye interaction. Solvents with high β and DN values show strong solvent–CyD interactions, yielding weak CyD–dye interactions. The interactions between methyl-β-CyD

and the dye can be adjusted by changing both the acid and basic properties of the medium. In solvents such as dichloromethane and chloroform, that exhibit very low a and β values, CyD–dye interactions became so strong that association complexes formed through hydrogen bonding could be detected.

This model fails if the solvent considered is water. Spectroscopic data involving **2** and the CyD in water revealed the formation of an inclusion complex, presenting a 1 : 1 stoichiometric ratio. The fact that dye **1** presents a bathochromic shift in its solvatochromic band when the CyD is added also suggests the inclusion of this dye in the CyD. The data set that registered bathochromic shifts for the two solvatochromic dyes indicates that the cavity of the CyD is hydrophobic. Attempts have been made by many authors to establish the polarity of the CyD cavity.[2] Although the hydrophobic character of the CyD cavity is well known,[1–4] the conclusions drawn from the experimental data are not in agreement with regard to the comparison of the cavity microenvironment with that of a pure solvent.[2,19] This is because the compounds used have different structures and interact in a different manner with the CyD cavity, thus reporting different micropolarities. Therefore, this study shows, through the use of probes **1** and **2** that the polarity of the cavity of methyl-β-CyD is lipophilic, but the imprecision regarding the effective location of the dye in the cavity makes the definition of an exact 'value' for this polarity difficult. Further research should be carried out with other classes of dyes in order to get a better and more profound understanding regarding this question.

Experimental

Materials

Methyl-β-CyD was kindly delivered by Wacker Chemie GesmbH (Germany) and used as received [Cavasol® W7M batch 71T023: degree of substitution per anhydro glucose unit of 1.8 (NMR), 0.20% of unsubstituted cyclodextrin, volatile organics < 0.20% (GC) and chloride content 0.60% (titration)]. Karl Fischer titrations were performed in a 0.02 mol dm^{-3} methyl-β-CyD solution in acetonitrile. These experiments demonstrated the presence of 10% (wt) of water in the CyD. All solvents were HPLC grade and were purified following methodologies described in the literature.[31] Deionized water was used in all measurements. This solvent was boiled and bubbled with nitrogen and kept under nitrogen atmosphere to avoid the presence of carbon dioxide. Dye **1** was synthesized as described previously.[32] Dye **2** was synthesized according to the method described in the literature,[33] recrystallized three times from hot water and dried under vacuum.

UV-vis spectra of the dyes in the solutions containing methyl-β-CyD

UV-vis measurements were performed with a Varian Cary Bio 50 spectrophotometer at 25 °C, with a precision of 0.1 nm, using a 1 cm quartz square cuvette. The maxima on the UV-vis spectra (λ_{max}) were calculated from the first derivative of the absorption spectrum. The λ_{max} values thus obtained were transformed into E_T(dye) values, given in kJ mol^{-1} according to the expression $E_T(\text{dye}) = 4.184 \times 28590/\lambda_{max}$.

A general procedure was used to take spectral measurements of the dye solutions in the HBD and HBA solvents in the presence of methyl-β-CyD. Dye solutions (1.0×10^{-4} and 4.7×10^{-5} mol dm^{-3} for **1** and **2**, respectively) were prepared in the solvent using a 50 cm^3 volumetric flask. A part of this solution was used in the preparation of a 0.04 mol dm^{-3} CyD stock solution in a 25 cm^3 volumetric flask. Solutions with different CyD concentrations were then prepared in 5 cm^3 volumetric flasks by the mixture of appropriate volumes from the two stock solutions and the UV-vis spectra were collected. The experiments using water were performed in buffered medium in order to avoid the protonation of the dye, the final pH being adjusted to 8.0.

The experiments with $CHCl_3$ and CH_2Cl_2 as solvents were performed with the preparation of the two stock solutions, one with dye **1** and the other containing **1** with methyl-β-CyD (0.04 mol dm^{-3}), as described in the previous paragraph. These solutions were sealed with a rubber septum and were kept under a stream of nitrogen. Then, 1.5 cm^3 of the dye solution were transferred to a 1.0 cm quartz square cuvette sealed with a rubber septum. A stream of nitrogen was passed through the cuvette and small amounts (0.100 cm^3) from the CyD stock solution were added with a microsyringe. The UV-vis spectrum was collected after each addition.

Measurements of association constants

The spectral data obtained in water with dye **2** and in chloroform and dichloromethane with dye **1** were analyzed according to the treatment described by Connors,[19,34] with eqn. (1),

$$\Delta A/b = [\text{dye}]K_{11}\Delta\varepsilon_{11}[\text{CyD}]/(1 + K_{11}[\text{CyD}]) \quad (1)$$

where b is the path length, [dye] is the dye concentration, $\Delta\varepsilon_{11}$ is difference in molar absorptivities of the complexed and free dye and K_{11} is the association constant for a 1 : 1 complex formation. ΔA is the change in absorbance at a fixed wavelength for the solution containing the dye when the concentration of the CyD changes from zero to [CyD]. The Benesi–Hildebrand equation [eqn. (2)] and the Scott equation [eqn. (3)] were used in order to estimate the initial values of $\Delta\varepsilon_{11}$ and K_{11} to be used in eqn. (1).[34]

$$b/\Delta A = 1/[\text{dye}]K_{11}\Delta\varepsilon_{11}[\text{CyD}] + 1/[\text{dye}]\Delta\varepsilon_{11} \quad (2)$$

$$b[\text{CyD}]/\Delta A = [\text{CyD}]/[\text{dye}]\Delta\varepsilon_{11} + 1/[\text{dye}]K_{11}\Delta\varepsilon_{11} \quad (3)$$

Calculations

The association constants were calculated using the ORIGIN 5.0 program.

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References

1 (*a*) M. L. Bender and M. Komiyama, *Cyclodextrin Chemistry*, Springer-Verlag, Berlin, 1978; (*b*) M. V. Rekharsky and Y. Ynoue, *Chem. Rev.*, 1998, **98**, 1875.
2 K. A. Connors, *Chem. Rev.*, 1997, **97**, 1325.
3 J. Szejtli, in *Inclusion Compounds*, ed. J. L. Atwood, J. E. D. Davies and D. D. MacNicol, Academic Press, London, 1984, Vol. 3, p. 331.
4 H. Dugas, *Bioorganic Chemistry: A Chemical Approach to Enzyme Action*, 3rd edn, Springer-Verlag, New York, 1996, p. 345.
5 M. Komiyama and M. L. Bender, *J. Am. Chem. Soc.*, 1978, **100**, 2259.
6 V. T. D'Souza and M. L. Bender, *Acc. Chem. Res.*, 1987, **20**, 146.
7 (*a*) S. Li and W. C. Purdy, *Chem. Rev.*, 1992, **92**, 1457; (*b*) C. Bicchi, A. D'Amato and P. Rubiolo, *J. Chromatogr., A.*, 1999, **843**, 99.
8 B. Chankvetadze, G. Endresz and G. Blaschke, *Chem. Soc. Rev.*, 1996, **25**, 141.
9 C. Reichardt, *Solvents and Solvent Effects in Organic Chemistry*, 2nd edn, VCH, Weinheim, 1988, ch. 6 and 7.
10 P. Suppan and N. Ghoneim, *Solvatochromism*, 1st edn, Royal Society of Chemistry, Cambridge, 1997, ch. 3.
11 C. Reichardt, *Chem. Rev.*, 1994, **94**, 2319.
12 Y. Marcus and Y. Migron, *J. Phys. Chem.*, 1991, **95**, 400.
13 (*a*) L. G. S. Brooker, C. H. Keyes, R. H. Sprague, R. H. Van Dyke, G. Van Zandt, F. L. White, H. W. J. Cressman and S. G. Dent, *J. Am. Chem. Soc.*, 1951, **73**, 5332; (*b*) L. G. S. Brooker, C. H. Keyes and D. W. Heseltine, *J. Am. Chem. Soc.*, 1951, **73**, 5350; (*c*) H. G. Benson and J. N. Murrell, *J. Chem. Soc., Faraday Trans. 2*, 1972, **68**, 137; (*d*) A. Botrel, A. Le Beuze, P. Jaques and H. Strub, *J. Chem. Soc.,*

Faraday Trans. 2, 1984, **80**, 1235; (*e*) P. Jacques, *J. Phys. Chem.*, 1986, **90**, 5535; (*f*) J. Catalan, E. Mena, W. Meutermans and J. Elguero, *J. Phys. Chem.*, 1992, **96**, 3615; (*g*) J. O. Morley, R. M. Morley, R. Docherty and M. H. Charlton, *J. Am. Chem. Soc.*, 1997, **119**, 10192; (*h*) J. O. Morley, R. M. Morley and A. L. Fitton, *J. Am. Chem. Soc.*, 1998, **120**, 11479.

14 See for instance: (*a*) V. Gageiro, M. Aillón and M. C. Rezende, *J. Chem. Soc., Faraday Trans. 2*, 1992, **88**, 201; (*b*) C. Reichardt, S. Asharin-Fard and G. Schäfer, *Chem. Ber.*, 1993, **126**, 143; (*c*) S. P. Zanotto, M. Scremin, C. Machado and M. C. Rezende, *J. Phys. Org. Chem.*, 1993, **6**, 637; (*d*) C. Machado, M. G. Nascimento, M. C. Rezende and A. E. Beezer, *Thermochim. Acta*, 1999, **328**, 155.

15 (*a*) K. Dimroth and C. Reichardt, *Z. Anal. Chem.*, 1966, **215**, 344; (*b*) Z. B. Maksimovic, C. Reichardt and A. Spiric, *Z. Anal. Chem.*, 1974, **270**, 100; (*c*) R. D. Skwierczynski and K. A. Connors, *J. Chem. Soc., Perkin Trans. 2*, 1994, 467; (*d*) M. Rosés, C. Ràfols, J. Ortega and E. Bosch, *J. Chem. Soc., Perkin Trans. 2*, 1995, 1607; (*e*) E. Bosch, M. Rosés, K. Herodes, I. Koppel, I. Leito, I. Koppel and V. Taal, *J. Phys. Org. Chem.*, 1996, **9**, 403; (*f*) K. Herodes, I. Leito, I. Koppel and M. Rosés, *J. Phys. Org. Chem.*, 1999, **12**, 109; (*g*) E. Humeres, R. J. Nunes, V. G. Machado, M. D. G. Gasques and C. Machado, *J. Org. Chem.*, 2001, **66**, 1163; (*h*) K. Herodes, J. Koppel, C. Reichardt and I. A. Koppel, *J. Phys. Org. Chem.*, 2003, **16**, 626.

16 (*a*) D. C. da Silva, I. Ricken, M. A. R. Silva and V. G. Machado, *J. Phys. Org. Chem.*, 2002, **15**, 420; (*b*) T. Bevilaqua, D. C. da Silva and V. G. Machado, *Spectrochim. Acta, Part A*, 2004, **60**, 951.

17 (*a*) K. A. Zachariasse, N. Van Phuc and B. Kozankiewicz, *J. Phys. Chem.*, 1981, **85**, 2676; (*b*) L. P. Novaki and O. A. El Seoud, *Langmuir*, 2000, **16**, 35; (*c*) E. B. Tada, L. P. Novaki and O. A. El Seoud, *Langmuir*, 2001, **17**, 652; (*d*) N. O. Mchedlov-Petrossyan, N. A. Vodolazkaya and C. Reichardt, *Colloids Surf., A*, 2002, **205**, 215.

18 See for instance: (*a*) F. V. Bright, T. L. Keimig and L. B. McGowan, *Anal. Chim. Acta*, 1985, **175**, 189; (*b*) G. C. Catena and F. V. Bright, *Anal. Chem.*, 1989, **61**, 905; (*c*) K. M. Tawarah and A. A. Wazwaz, *J. Chem. Soc., Faraday Trans.*, 1993, **89**, 1729; (*d*) N. J. Crane, R. C. Mayrhofer, T. A. Betts and G. A. Baker, *J. Chem. Educ.*, 2002, **79**, 1261; (*e*) M. A. El-Kemary, H. S. El-Gezawy, H. Y. El-Baradie and R. M. Issa, *Spectrochim. Acta, Part A*, 2002, **58**, 493; (*f*) S. Panja and S. Chakravorti, *Spectrochim. Acta, Part A*, 2002, **58**, 113; (*g*) M. A. El-Kemary and H. S. El-Gezawy, *J. Photochem. Photobiol. A*, 2003, **155**, 151.

19 K. A. Connors, M. J. Mulski and A. Paulson, *J. Org. Chem.*, 1992, **57**, 1794.

20 M. J. Kamlet, J.-L. M. Abboud, M. H. Abraham and R. W. Taft, *J. Org. Chem.*, 1983, **48**, 2877.

21 Y. Marcus, *J. Phys. Chem.*, 1987, **91**, 4422.

22 Y. Marcus, *Ion Solvation*, John Wiley and Sons, New York, 1985.

23 (*a*) V. Gutmann, *Coord. Chem. Rev.*, 1967, **2**, 239; (*b*) Y. Marcus, *J. Solution Chem.*, 1984, **13**, 599.

24 Y. Marcus, *J. Solution Chem.*, 1991, **20**, 929.

25 J. Ortega, C. Ràfols, E. Bosch and M. Rosés, *J. Chem. Soc., Perkin Trans. 2*, 1996, 1497.

26 O. S. Wolfbeis and M. A. Kessler, *Chem. Phys. Lipids*, 1989, **50**, 51.

27 S. J. Davidson and W. P. Jencks, *J. Am. Chem. Soc.*, 1969, **91**, 225.

28 C. A. Coleman and C. J. Murray, *J. Org. Chem.*, 1992, **57**, 3578.

29 C. Laurence, P. Nicolet, M. Lucon, T. Dalati and C. Reichardt, *J. Chem. Soc., Perkin Trans. 2*, 1989, 873.

30 (*a*) J. G. Dawber and R. A. Williams, *J. Chem. Soc., Faraday Trans. 1*, 1986, **82**, 3097; (*b*) J. G. Dawber, J. Ward and R. A. Williams, *J. Chem. Soc., Faraday Trans. 1*, 1988, **84**, 713.

31 B. S. Furniss, A. J. Hannaford, P. W. G. Smith and A. R. Tatchell, *Vogel's Textbook of Practical Organic Chemistry*, 5th edn, Longman, London, 1989.

32 M. C. Rezende and C. M. Radetski, *Quim. Nova*, 1988, **11**, 353; M. C. Rezende and C. M. Radetski, *Chem. Abstr.*, 1989, **111**, 8876w.

33 M. J. Minch and S. S. Shah, *J. Chem. Educ.*, 1977, **54**, 709.

34 K. A. Connors, *Binding Constants: the Measurement of Molecular Complex Stability*, Wiley-Interscience, New York, 1987, ch. 4.