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Complexation of Synthetic Organic Dye Dapoxyl with cyclodextrins studied by fluorescence spectroscopy

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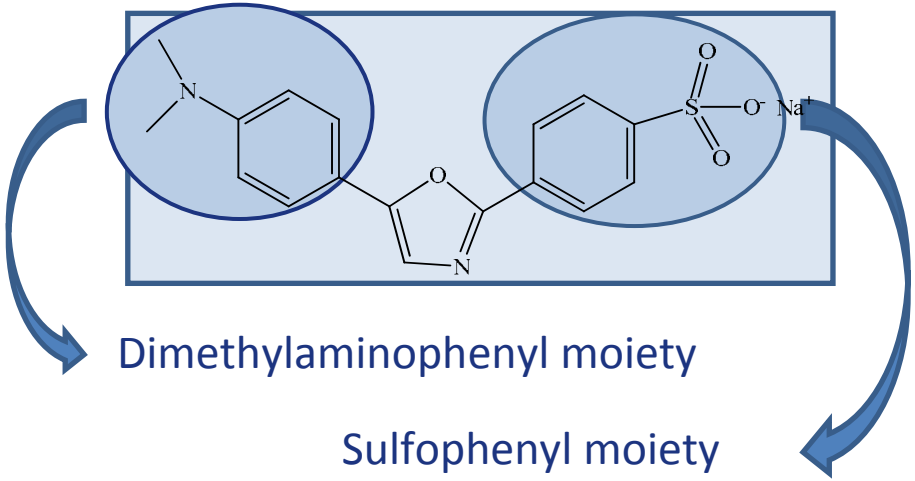
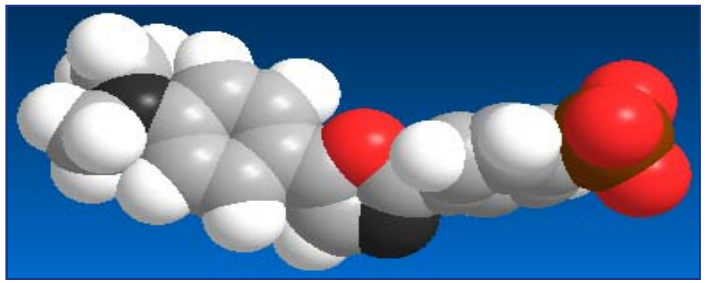
Abstract: Cyclodextrins (CDs) are natural-occurring oligomers with a hydrophobic cavity which allow them to form inclusion complexes in water with a variety of organic molecules. These complexes are generally stabilized by van der Waals and hydrophobic interactions, although specific host-guest interactions can also play an important role.[1] In addition, geometrical and orientational requirements of both the guest and the host may control the association process, whereas the dissociation rate is determined by the strength of the interactions.[2] Therefore, a change of the cavity size or the rigidity of the host has dramatic effects on both the association and the dissociation rate constants and in consequence on the stability of the complexes.[3] Moreover, for a certain guest, stoichiometry and geometry of the inclusion complexes may also be much dependent on the host cavity size.[4] In this work we study the effects of CD cavity size on the stoichiometry, stability and structure of the complexes formed between the synthetic organic probe Dapoxyl and three natural cyclodextrins (α-CD, β-CD and γ-CD) differing in the size of their inner cavity, using steady-state and time-resolved fluorescence techniques. The ditopic structure of this fluorophore together with its high sensitivity to the surroundings make it a useful model molecule to study the geometrical effects on the complexation. Complexation provokes strong changes in the fluorescent properties of Dapoxyl with a large blue shift of its emission spectrum and a great increase of the fluorescence quantum yield. The differences observed for the three CDs regarding stoichiometry, stability and fluorescence properties of the complexes are discussed on the basis of a size-selective complexation of the Dapoxyl.

Introduction: Dapoxyl sodium sulfonate

Fluorescent probe:

2-(4'-Sulfophenyl)-5-(4''-dimethylaminophenyl)oxazole, sodium salt (DAP)

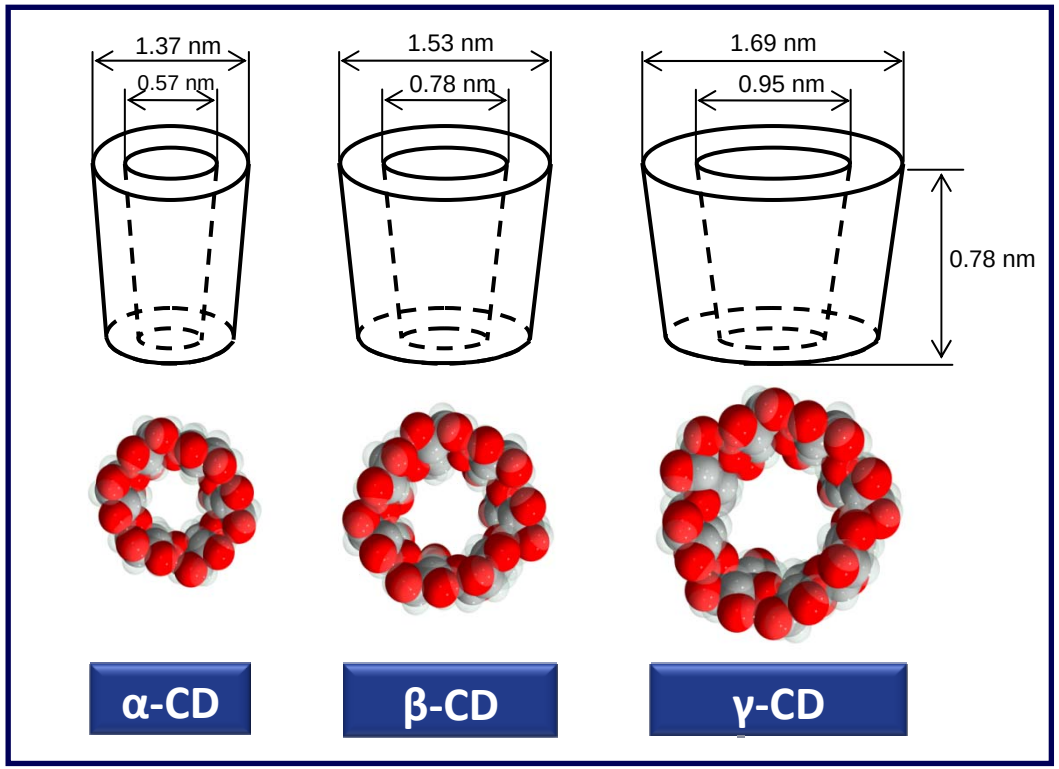


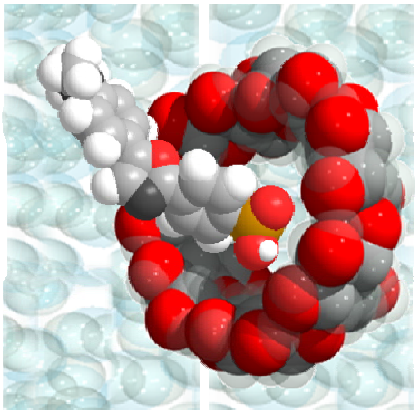


› **Higher sensitivity to the surroundings than other probes:** Fluorescence maximum and fluorescence quantum yield very sensitive to solvent polarity

› Other interesting properties: long emission wavelengths, high extinction coefficients and large Stokes shifts

Introduction: Cyclodextrins





Inclusion complexes are simple and useful models for the investigation of host-guest interactions in supramolecular systems

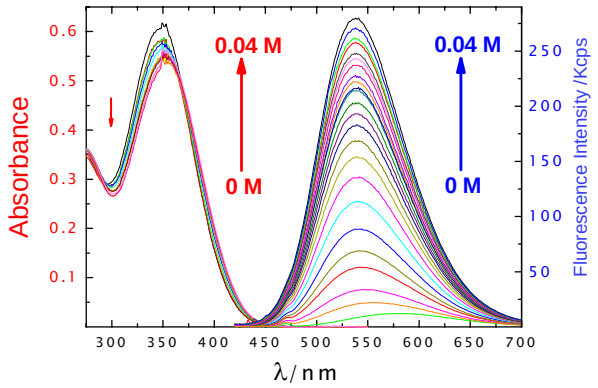
› Natural-ocurring cyclic oligosaccaharides obtained by enzymatic conversion of starch

› With a toroidal shape, CDs have a highly hydrophobic central cavity which allows them to form inclusion complexes with many organic substrates

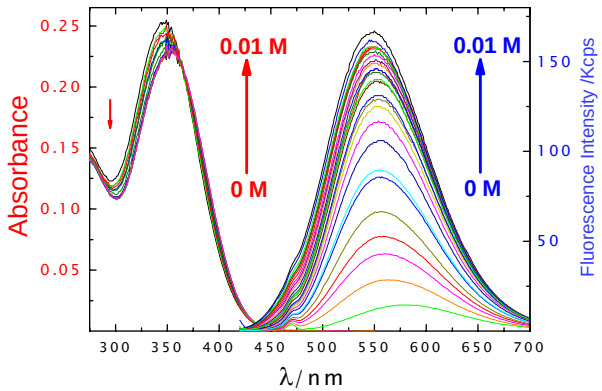
› Many applications: Pharmaceutical industry (controlled release), food technology, chemical industry, agricultural and analytic chemistry, …

Methodology and results

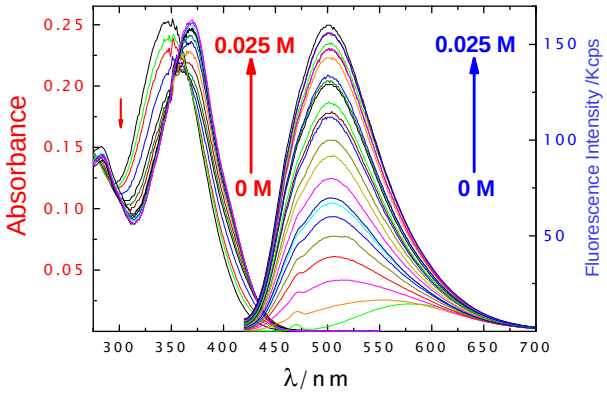
1.- Absorption and steady-state fluorescence titrations



α-CD



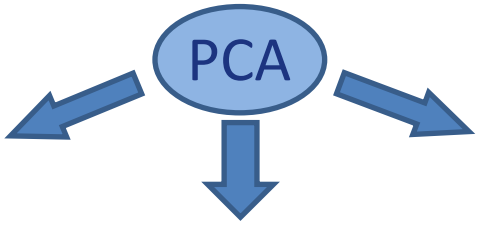
β-CD



γ-CD

The figures show the variations of the absorption and fluorescence emission spectrum of Dapoxyl with increasing concentrations of the three CDs. Small but systematic red shifts are observed in the absorption spectra. Emission spectra show strong blue shifts and huge increase of the fluorescence intensity.

2.- Determination of the number of fluorescent species: Analysis of the series of absorption and emission spectra at different CD concentrations using Principal Component Analysis (PCA) yield the minimal number of species contributing to the experimental spectra.



α-CD: 2 species

β-CD: 3 species

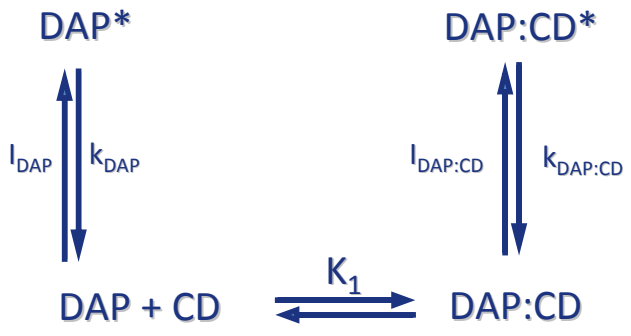
γ-CD: 3 species

3.- Proposal of the complexation mechanisms and fit functions

Taking into account the results of PCA, the following mechanisms are proposed to explain the behaviour of Dapoxyl in the presence of the different cyclodextrins. Only complexation equilibria in the ground state are considered since the association and dissociation processes are too slow to compete with the deactivation.

α-CD: 2 species

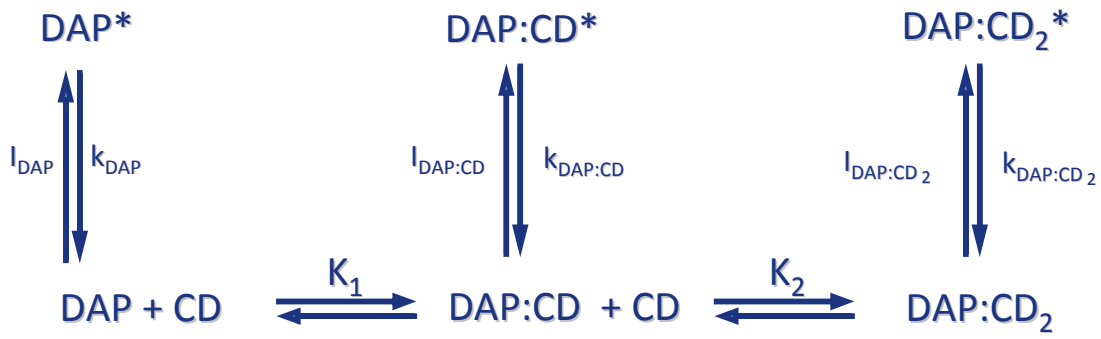
Complexation mechanism 1:1



$$P(\lambda) = \frac{a(\lambda) + b(\lambda) \cdot K_1 \cdot [CD]_0}{1 + K_1 \cdot [CD]_0} \quad P = A \text{ or } F$$

β-CD and γ-CD: 3 species

Complexation mechanism 1:1 + 1:2



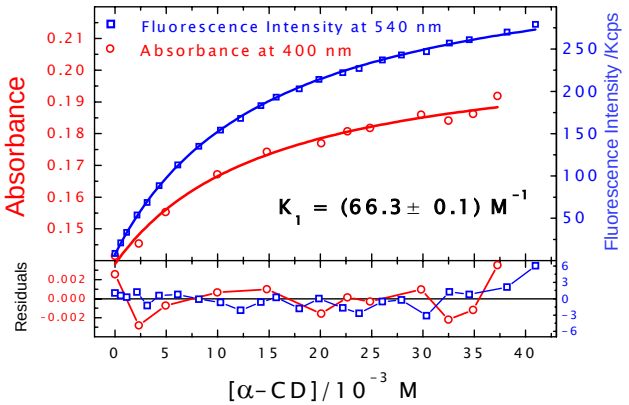
$$P(\lambda) = \frac{a(\lambda) + b(\lambda) \cdot K_1 \cdot [CD]_0 + c(\lambda) \cdot K_1 \cdot K_2 \cdot [CD]_0^2}{1 + K_1 \cdot [CD]_0 + K_1 \cdot K_2 \cdot [CD]_0^2} \quad P = A \text{ or } F$$

Methodology and results

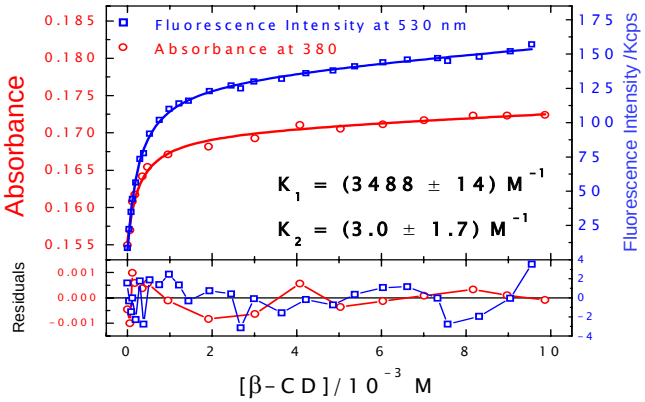
4.- Global Analysis (GA) (global fit to all wavelengths)

Global analysis (GA) of the series of emission and absorption spectra as datasets yield precise values of the association equilibrium constants K_1 in the case of α-CD and K_1 and K_2 for β-CD and γ-CD.

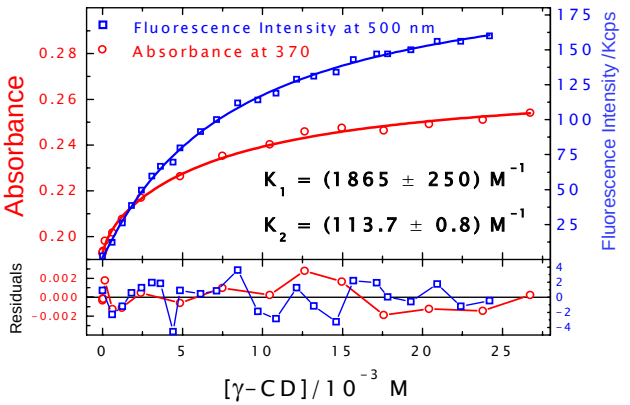
α-CD: 1:1 Complexation



β-CD: 1:1 + 1:2 Complexation



γ-CD: 1:1 + 1:2 Complexation



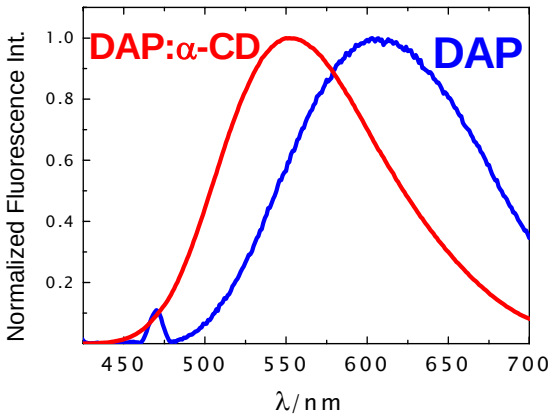
System	DAP + α-CD	DAP + β-CD	DAP + γ-CD
Species	**2**: (DAP + DAP:α-CD)	**3**: (DAP + DAP:β-CD + DAP:β-CD_2)	**3**: (DAP + DAP:γ-CD + DAP:γ-CD_2)
K_1 /M^{-1}	66.3 ± 0.1	3488 ± 14	1865 ± 250
K_2 /M^{-1}	-	3.0 ± 1.7	113.7 ± 0.8

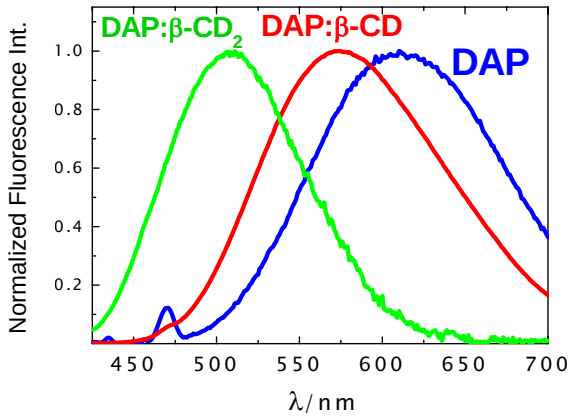
The stability constants of the complexes are very different for the three CDs showing the effect of the cavity sizes in the association processes as well as in the strength of the host-guest interactions.

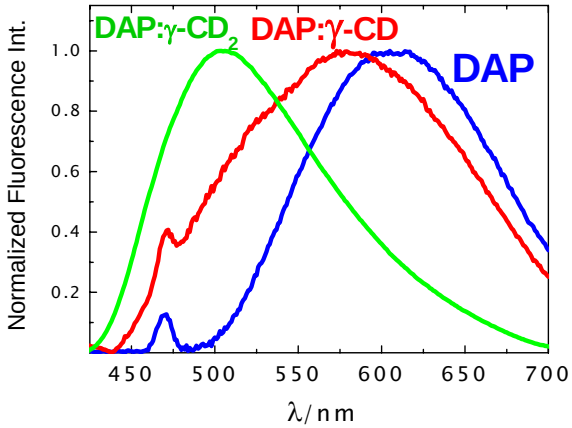
Methodology and results

5.- Obtaining the pure spectra (by means of PCGA: PCA + GA)

GA yields also the pure absorption and emission spectra of the free Dapoxyl and the Dapoxyl bound to the different CDs forming 1:1 (α, β and γ-CD) and 1:2 (only β and γ-CD) complexes. The figures show the normalized emission spectra. Complex formation causes a blue shift which is larger for the 1:2 complexes than for the 1:1 complexes. The broad spectrum obtained for the DAP:γ-CD complex suggests that two different types of 1:1 complexes are formed with γ-CD.







When calculating the fluorescence quantum yield from the pure spectra, we observe that it largely increases with the formation of the complexes of DAP:α-CD (1:1 complex), DAP:β-CD (1:1 and 1:2 complexes) and DAP:γ-CD (1:2 complex), but no change is observed in the 1:1 complex of DAP:γ-CD.

DAP
Φ =0.03

Species	Φ
DAP:α-CD	0.39

Species	Φ
DAP:β-CD	0.22
DAP:β-CD$_2$	0.51

Species	Φ
DAP:γ-CD	0.02
DAP:γ-CD$_2$	0.13

6.- Fluorescence lifetime measurements (TCSPC)

Lifetime measurements were carried out with picosecond resolution in order to obtain the lifetimes of free Dapoxyl and of the different complexes formed with the three CDs. Taking the lifetime values and the fluorescence quantum yields obtained earlier, we calculated the rate constants of the radiative and nonradiative deactivation processes (k_r and k_{nr}, respectively).

A biexponential decay is observed for free Dapoxyl in aqueous solution and this result is interpreted by the existence of two emissive states of the probe, being the main lifetime of 0.450 ns.

	Species	τ / ns	k_r/ns^{-1}	k_{nr}/ns^{-1}
	DAP	0.080 / 0.450	0.07	2.15
α	DAP:α-CD	3.90	0.10	0.15
β	DAP:β-CD	2.40	0.10	0.33
	DAP:β-CD_2	7	0.07	0.07
γ	DAP:γ-CD	1.37 / 3.67	-	-
	DAP:γ-CD_2	7.85	0.02	0.11

› The lifetime of the complex DAP:α-CD is much longer than that of free DAP due to a great decrease of the nonradiative rate constant.

› Nonradiative processes are also much less effective in the complexes with β-CD than in free DAP, specially in DAP:β-CD_2.

› Two lifetimes are obtained for DAP:γ-CD, confirming that this CD forms two different 1:1 inclusion complexes with each of the guest moieties. The lifetime of the 1:2 complex is very similar to that formed by β-CD.

Conclusions

› The high sensitivity of Dapoxyl to the environment makes it a useful probe to study the formation of inclusion complexes by CDs.

› The two different moieties of the Dapoxyl molecule can be included into the cavity of the CDs, allowing one to study the possible specific interactions.

› The type and stability of the distinct formed complexes will be determined by the inner cavity size of the CD.

› The large increase of the fluorescence quantum yield of Dapoxyl in apolar media allows us to study the dynamics in future investigations by means of fluorescence correlation spectroscopy (FCS).

› Further studies will be performed to determine the structures of the different complexes formed between Dapoxyl and the three cyclodextrins.

References

[1] Reija, B. et al. J. Phys. Chem. 109 (2005) 1364.

[2] Al-Soufi, W. et al. J. Am. Chem. Soc. 127 (2005) 8775.

[3] Al-Soufi, W. et al. ChemPhysChem 9 (2008) 1819.

[4] Bordello, J. et al. ChemPhysChem 10 (2009) 931.

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