Spectroscopic and self-association behavior of a porphyrin-β-cyclodextrin conjugate

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A novel porphyrin–cyclodextrin conjugate, 5[4-(6-*O*-β-cyclodextrin)-phenyl],10,15,20-tri(4-hydroxyphenyl)-porphyrin (**CD-THPP**), has been synthesized in good yield (30%) starting from 6-*O*-tosyl-β-cyclodextrin using 5,10,15,20-tetra(4-hydroxyphenyl)porphyrin (**p-THPP**) as a nucleophile. The spectroscopic properties of **CD-THPP** have been investigated by combining NMR, circular dichroism, UV-vis absorption, fluorescence and laser flash photolysis techniques, allowing us to gaining insights into the self-association mode of the conjugate. In neat methanol, **CD-THPP** is present in a monomeric form and exhibits spectroscopic features virtually identical to those of the model compound **p-THPP**. The role of the CD macrocycle is essential in differentiating the behavior of the conjugate with respect to the non-modified porphyrin in the mixture water : methanol, 9 : 1 (v/v). Under these conditions, **CD-THPP** shows a marked tendency to self-arrange as a dimer, whose formation is strongly encouraged by stacking interactions between the aromatic rings, in addition to intermolecular hydrophobic porphyrin–cyclodextrin interactions. This supramolecular species exhibits a strong exciton coupling and, contrary to the unmodified **p-THPP**, shows a good response to light excitation, as confirmed by the considerable fluorescence emission and triplet–triplet transient absorption. It is also demonstrated that incorporation of suitable guests into the CD cavity triggers a new rearrangement of the dimer with a higher degree of overlap between the porphyrin chromophores, facilitating the occurrence of intramolecular photoinduced electron transfer between the guest and the porphyrin unit.

Introduction

The conjugation of molecules with different properties is a very attractive approach in the building of multifunctional systems devoted to applications in different fields. Cyclodextrins (CDs) are water soluble cyclic oligosaccharides that have been extremely appealing for a long time to investigators in both pure and applied fields.[1] A number of CD-based conjugates with amines,[2] amino acids,[3] peptides[4] and aromatic systems[5] have been reported over the years. Fascinating compounds, including enzyme mimics,[2–7] abiotic receptors,[6–9] fluorescence indicators[10–14] and molecular actuators,[15] can be obtained by exploiting the appropriate combination of the hydrophobic nature and different size of the CD cavity with the specific features of the *ad hoc*-attached molecules.

Porphyrins and their metal complexes represent a family of heavily investigated molecules by virtue of their excellent spectroscopic, photochemical and electrochemical properties, as well as their tendency to form aggregates of different order and structure. Commonly, such self-assembled species can be classified as being of H- or J-type on the basis of the relative orientations of the induced transition dipoles of the porphyrins, either *head-to-head* or *head-to-tail*, respectively.[16] The literature is very rich in examples of porphyrin-based conjugates with various classes of molecules, including other porphyrins (mono- and heterodimers),[17] macrocycles,[7–10] peptides,[18] calixarenes[19,20] and carbohydrates.[11] These chromophoric systems represent effective models of photosynthetic electron transfer,[21] singlet or triplet energy transfer[22] and inter-chromophore interactions,[23] and are well-suited to a large variety of potential applications in many fields of technological interest, spanning from photoconductors and chemical sensors[24] to enzyme mimetics and chemical recognition.[6]

A number of inclusion complexes of CDs/porphyrins have been characterized,[8,9,12] and the related modification of photochemical and aggregation behavior has also been reported. The use of amino CDs as hosts of anionic porphyrins has also proved to be a valuable strategy for the improvement of binding efficiency by exploiting the electrostatic interaction.[25] In other cases, porphyrin assemblies with high stability constants have been achieved by using dimers of CDs.[26] On these bases, the conjugation of CDs with porphyrins is of particular relevance in light of the appropriate combination of binding and recognition ability of the CD unit with the photo and/or redox properties of the porphyrin chromophore. These systems have attracted considerable interest in recent years for studying binding,[27] catalytic[28] and photoinitiated processes.[29] In the field of biomimetic chemistry, some models of biological

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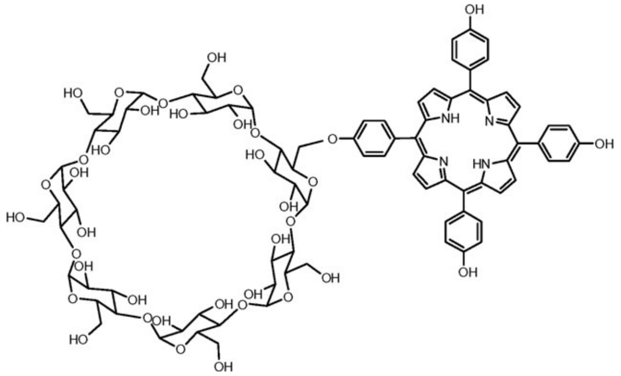


Fig. 1 The chemical structure of **CD-THPP**.

systems, such as cytochrome P-450,[15] O_2 carriers[30,31] and electron transfer systems,[32] have also been also reported. Furthermore, the CD–porphyrin conjugates have been used as hosts of porphyrins to form heteroporphyrin arrays in aqueous solution[33] and to build miniaturized devices.[13,14] Recently, Carofiglio *et al.* have reported on the supramolecular properties of a conjugate of β-CD with an anionic porphyrin,[34] elegantly pointing out the key role of the CD unit in determining the peculiar association mode. On these grounds, in this work, we considered it useful to investigate the spectroscopic and self-association behavior of a novel conjugate of β-CD with a non-ionic porphyrin, 5[4-(6-*O*-β-cyclodextrin)-phenyl],10,15,20-tri(4-hydroxyphenyl)porphyrin (**CD-THPP**) (Fig. 1), with the aim of providing an additional contribution to the general picture of CD–porphyrin conjugate systems. The study was carried out in two different solvents, dependent on pH, and in the presence of either suitable guest or host molecules by combining NMR, circular dichroism, UV-vis absorption, fluorescence emission and laser flash photolysis techniques.

Experimental

Materials and general procedures

β-Cyclodextrin (Fluka), anhydrous *N*,*N*-dimethylformamide (Aldrich) and 5,10,15,20-tetra(4-hydroxyphenyl)porphyrin (**p-THPP**, Porphyrin Systems) were used without further purification. Anthraquinone-2-sulfonate (**AQS**) and 1-adamantanol (**ADM**) were purchased from Sigma-Aldrich. Thin Layer Chromatography (TLC) was carried out on silica gel plates (Merck 60-F254). Merck Lichroprep RP-8 (40–60 μm) was used for reversed phase column chromatography. CD derivatives were detected on TLC by UV and by an anisaldehyde test. Analytical and preparative HPLC were carried out on a Varian PrepStar instrument equipped with a ProStar 330 photodiode array detector. Preparative (5 μm, 22 × 250 mm) and analytical (5 μm, 2.0 × 250 mm) Econosphere ODS (Alltech) columns were used.

Synthesis of CD-THPP. 6-Tosyl β-cyclodextrin[35] (0.800 g, 0.622 mmol) was added to a dry DMF solution (10 ml) of **p-THPP** (0.500 g, 0.738 mmol) and Cs_2CO_3 (0.240 g, 0.738 mmol). The reaction mixture was stirred under nitrogen at 140 °C for 1 d. The solvent was evaporated to dryness *in vacuo*, the solid washed with MeOH and purified by preparative HPLC (linear gradient 0.10 M $AcNH_4$ ($pH = 5$) $\rightarrow$ CH_3CN). The appropriate fractions were concentrated to give **CD-THPP**. The purity of the product was verified by analytical HPLC. TLC: $R_f = 0.76$ (PrOH/H_2O/AcOEt/NH_3 5 : 3 : 1 : 2), yield: 30%.

ESI-MS: $m/z = 1795.3$ $[M + H]^+$, 1818.2 $[M + Na]^+$, 909 $[2M + 2Na]^{2+}$.

^1H NMR (CD_3OD, 500 MHz). A notation system, in which the glucose rings are named counter-clockwise A, B, C, D, E, F and G, and viewed from the primary hydroxyl side, is adopted. δ (/ppm): 8.70 (s, 8H, β-pyrrole), 7.92 (d, 2H, phenyl H-2), 7.84 (m, 6H, phenyl H-2), 7.24 (d, 2H, phenyl H-3), 7.05 (m, 6H, phenyl H-3), 4.98 (d, 1H, H-1G), 4.96 (d, 1H, 1A), 4.84 (d, 1H, H-1B), 4.80 (m, 4H, other H-1), 4.60 (d, 1H, H-6A), 4.45 (dd, 1H, H-6′A), 4.13 (m, 1H, 5A), 3.94–3.60 (m, 25H, H-6,-5,-3, 4A), 3.51 (dd, 1H, H-2A), 3.50–3.30 (m, 13H, H-2,-4). Elemental analysis for $C_{86}H_{98}N_4O_{38}$: calc. C, 57.52; H, 5.50; N, 56.03; found: C, 56.99; H, 5.39; H, 55.67%.

Instrumentation

ESI-MS measurements were carried out using a LCQ DECA XP PLUS spectrometer equipped with an ESI source. Solutions were injected into the ion source at a flow rate of 10 μl min^{-1} using nitrogen as the drying gas. The mass spectrometer operated with a capillary voltage of 46 V and a capillary temperature of 200 °C, while the spray voltage was 4.3 kV.

Absorption and emission spectra were recorded with a Hewlett-Packard HP 8452 spectrophotometer and a Spex Fluorolog-2 (mod. F-111) spectrofluorimeter, respectively.

Absolute fluorescence quantum yields were calculated by using **p-THPP** in methanol as a secondary standard. In determining the relative fluorescence quantum yields at different excitation wavelengths, the relative number of absorbed photons was taken into account, and was considered to be the integral of the product of the absorbance and the relative photon intensity of the excitation light source. Circular dichroism spectra were recorded with a Jasco J-800 dichrograph. Calibration of the instrument was performed with a 0.06% solution of ammonium camphorsulfonate in water (molar circular dichroism $\Delta\varepsilon = 2.40$ at 290.5 nm). The spectral range between 200 and 700 nm was covered using quartz cells of 0.1 or 1 cm path lengths. Results are reported in terms of ε and $\Delta\varepsilon$.

^1H NMR spectra were recorded at 25 °C in CD_3OD on a Varian Inova 500 spectrometer at 499.883 MHz. ^1H NMR spectra were measured using standard pulse programs from the Varian library. In all cases, the length of the 90° pulse was *ca.* 7 μs. 2D experiments were carried out using 1 K data points, 256 increments and a relaxation delay of 1.2 s. T-ROESY spectra were obtained with a 300 ms spin-lock time. 2,2-Dimethyl-2-silapentane-5-sulfonate sodium salt DSS was used as an external standard.

Nanosecond laser flash photolysis setup. All samples were excited in a cell of 1 cm optical path length with the second harmonic ($\lambda_{exc} = 532$ nm) of a Nd–YAG Continuum Surelite II-10 laser system (pulse width 6 ns FWHM), and the excited solutions were analyzed at right angles using a mLFP-111 apparatus developed by Luzchem Research. The monitoring beam was supplied by a ceramic xenon lamp and delivered through quartz fiber optical cables. The laser pulse was probed by a fiber, which synchronized the mLFP system with a Tektronix TDS 3032 digitizer operating in the pre-trigger mode. The signals from a compact Hamamatsu photomultiplier were initially captured by the digitizer and then transferred to a personal computer, which controlled the experiments using Luzchem-developed software in the LabView 5.1 environment from National Instruments. The energy of the laser pulse was measured at each laser shot by a SPHD25 Scientech pyroelectric energy monitor. The solutions for the experiments in the absence of oxygen were deoxygenated by bubbling through a vigorous and constant flux of pure argon (previously saturated with solvent). The sample temperature was 295 ± 2 K. The energy of the laser pulse was measured at each shot with a SPHD25 Scientech pyroelectric meter.

Results

NMR spectroscopy

The ^{1}H NMR spectrum of **CD-THPP** measured in deuterated methanol is shown in Fig. 2. The 1D spectrum was assigned by COSY, TOCSY, T-ROESY and HSQC.

In the aromatic region, signals due to the porphyrin moiety are present; at 8.9 ppm, the large peak due to the methylenes of the pyrrole rings; at 7.8 and at 7.4 ppm, the signal of the phenyl bound to the CD, respectively, H-2 and H-3; and the other phenyl protons at 8.0 and 7.2 ppm. More groups of signals appear in the H-1 region. Functionalization breaks the symmetry of the cavity, and H-1G and H-1B, in addition to H-1A, can be identified on the spectrum by T-ROESY. Other signals due to the functionalized glucose ring are well evident on the spectrum at 4.60 ppm, the 6-A protons at 4.45 ppm and the 5A protons at 4.13 ppm.

The T-ROESY spectrum shows correlations between the aromatic region (phenyl H-3) and the H-6A protons of CD, in keeping with the proximity of the phenyl ring bound to the primary rim of the cavity. The intramolecular interaction of the porphyrin with the CD cavity can be excluded on the basis of CPK models. In fact, the porphyrin moiety is too rigidly bound to the cavity and does not possess sufficient flexibility to be intramolecular included. In keeping with this, other correlation peaks between the porphyrin and CD protons are evident in the spectrum, thus suggesting that the porphyrin ring is not included in the cavity, not even intermolecularly. This is further confirmed by the lack of changes to the ^{1}H NMR spectrum upon addition of an effective guest for the CD cavity, such as 1-adamantanol (**ADM**).[36] The very low solubility of **CD-THPP** in D_2O or a D_2O : CD_3OD mixture precluded ^{1}H NMR being undertaken in these solvents.

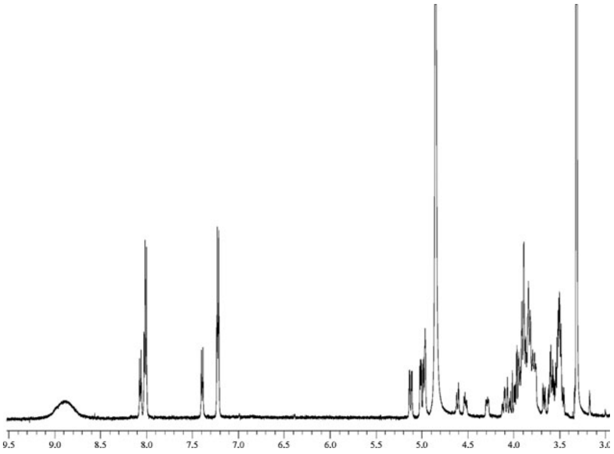


Fig. 2 ^{1}H NMR spectrum of **CD-THPP** (CD_3OD, 500 MHz).

Absorption and emission

Fig. 3 shows the absorption and fluorescence emission spectra of **CD-THPP** recorded in neat methanol. The absorption spectrum is characterized by the intense Soret band centered at 420 nm and weaker Q-bands with maxima at 519, 559 and 595 nm. The fluorescence spectrum consists of two emission bands with maxima at 653 and 717 nm. These spectroscopic features, as well as the value of the fluorescence quantum yield (Φ_f = 0.13), are virtually identical to those exhibited by **p-THPP**, chosen as model compound, in the same solvent[37] and are not affected by the presence of **ADM**. This finding reveals that the porphyrin chromophore retains its spectroscopic properties upon attachment of the CD macrocycle, ruling out (according to the NMR results) any intra- or intermolecular interactions between the two sub-units of **CD-THPP**.

Fig. 4a and b show the absorption and fluorescence spectra of **CD-THPP** alone, in the presence of **ADM** and, for comparison, the spectra of the model compound **p-THPP**, in a mixture water : methanol, 9 : 1 (v/v). The spectroscopic behaviors observed in this medium are considerably different from those observed in neat methanol. In fact, although no change in the λ_{max} values are noted, the Soret band of **CD-THPP** is significantly broadened and exhibits large hypochromicity (*ca.* 50%). These effects are much more pronounced in the case of the model compound **p-THPP**. Addition of **ADM** to solutions of the CD conjugate further increases both hypochromism and broadening of the Soret band. In particular, this latter becomes very similar to that of **p-THPP** (see inset Fig. 4a, for sake of clarity). The emissive properties better reflect the differences between **CD-THPP** and the model compound **p-THPP**. Indeed, whilst the fluorescence quantum yield of **p-THPP** drops drastically down, Φ_f = 0.0012 (*ca.* 2

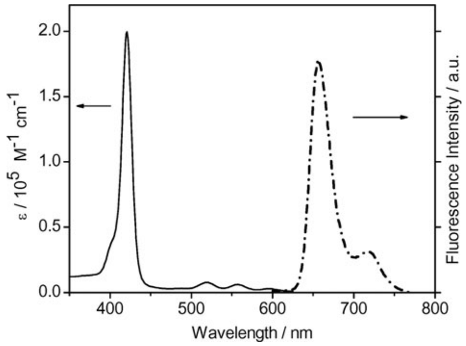


Fig. 3 Absorption (—) and emission (---) spectra of **CD-THPP** (1 μM) recorded in methanol solution. λ_{exc} = 420 nm.

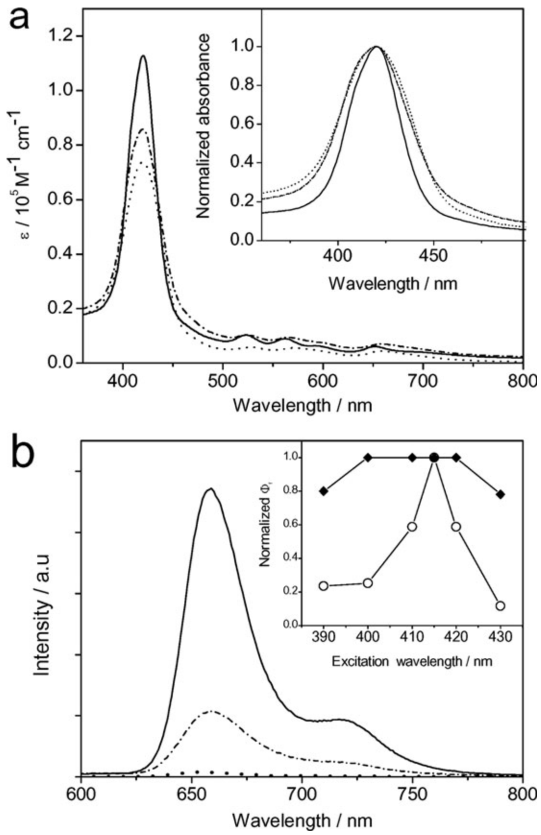


Fig. 4 Absorption (a) and fluorescence (b) spectra of **CD-THPP** (—), **CD-THPP** in the presence of 4 × 10^{-4} M **ADM** (-·-) and **p-THPP** in water : methanol, 9 : 1 (v/v) (···). The fluorescence spectra are corrected for the different amounts of photons absorbed by each sample at the excitation wavelength (λ_{exc} = 420 nm). Inset Fig. 4a: Normalized absorption of the Soret band region at an expanded scale. Inset Fig. 4b: Normalized wavelength dependence of the emission quantum yields of **CD-THPP** (♦) and **p-THPP** (○). [**CD-THPP**] = [**p-THPP**] = 1 μM.

order of magnitude smaller than in methanol), in the case of **CD-THPP** the presence of the CD sub-unit leads to a considerable revival of the emission. The fluorescence quantum yield of the CD conjugated is in fact Φ_f = 0.06, a value *ca.* 50-fold larger than that of the model compound and only two-fold small when compared to that exhibited in neat methanol. Moreover, it should be noted that while the weak emission of the **p-THPP** appears at the same λ_{max} as in methanolic solution, the fluorescence maximum of **CD-THPP** is red shifted of *ca.* 6 nm. Likewise the absorption, **ADM** has a strong effect also on the fluorescence properties of **CD-THPP**. More than 75% of quenching is in fact noted in the presence of this guest molecule.

The presence of the CD macrocycle in the structure of **CD-THPP** also has a profound effect on the excitation wavelength dependence of the fluorescence quantum yield. The results obtained are displayed in the inset of Fig. 4b where, for the sake of clarity, the data have been normalized due to the large difference in the absolute values of Φ_f between the CD conjugate and the model compound. Although quite small, the emission efficiency of **p-THPP** is remarkably dependent on the excitation wavelength. In contrast, the Φ_f of **CD-THPP** is independent of the excitation energy in the almost whole Soret region.

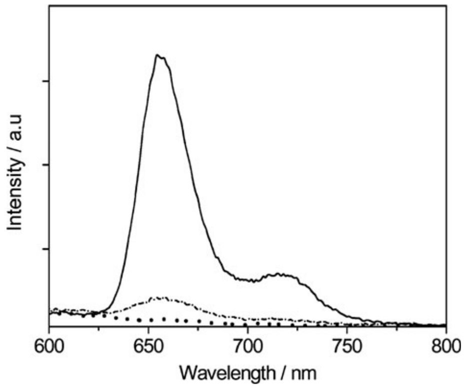


Fig. 5 Fluorescence spectra of **CD-THPP** (1 μM) alone (—), after addition of **AQS** 5 × 10^{-4} M (···) and after further addition of **ADM** 5 × 10^{-3} M (-·-) in water : methanol, 9 : 1 (v/v). λ_{exc} = 420 nm. The spectra are corrected for the different amounts of absorbed photons by each sample at the excitation wavelength.

The addition of a typical electron acceptor, such as anthraquinone-2-sulfonate (**AQS**), to the water : methanol solution of **CD-THPP** leads to a complete suppression of the fluorescence intensity (Fig. 5). However, it is interesting to note that further addition of **ADM** to this solution leads to some revival of the fluorescence emission, whose intensity becomes almost comparable to that obtained after adding **ADM** to a solution of **CD-THPP** in the absence of **AQS** (see Fig. 4b for comparison).

Circular dichroism spectra

The circular dichroism spectrum of **CD-THPP** (2.0 × 10^{-5} M) in MeOH is characterized by the absence of a signal corresponding to porphyrin absorption (data not shown). On the other hand, the circular dichroism spectrum recorded in water : methanol, 9 : 1 shows a strong exciton coupling, with the positive and negative bands peaking at 419 and 432 nm, respectively (Fig. 6).

This finding clearly suggests the proximity of porphyrin moieties under these experimental conditions.[38] It should also

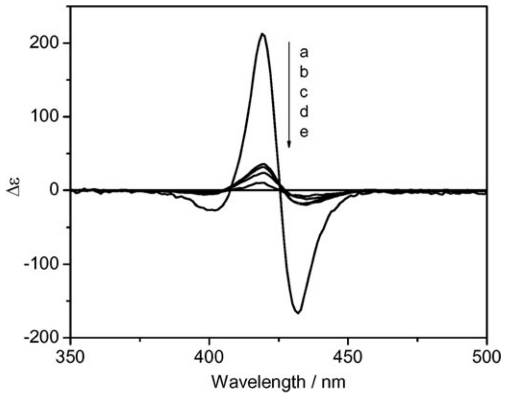


Fig. 6 Circular dichroism spectra of **CD-THPP** (1.6 × 10^{-5} M) alone (a) and in the presence of increasing amounts of **ADM** from 2.2 × 10^{-5} M to 3.7 × 10^{-4} M (b to e).

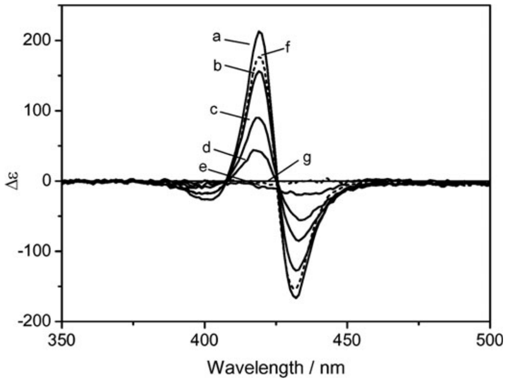


Fig. 7 Circular dichroism spectra of **CD-THPP** (1.5×10^{-5} M) at pH 7.0 (a), 8.4 (b), 9.0 (c), 9.5 (d), 12.0 (e), 4.5 (f) and 3.5 (g).

be noted that the Δε values of the circular dichroism spectrum did not depend on the concentration of **CD-THPP** over the wide range of concentrations investigated (7.0×10^{-7}–2.0×10^{-5} M), as well as by the addition of β-CD alone as a competitive host cavity, pointing to a very strong interaction between the porphyrin molecules. In contrast, the intense circular dichroism signal is strongly dependent on the presence of **ADM**, as evidenced by the reduction in intensity of the exciton coupling band with increasing molar ratio **ADM**/**CD-THPP** (Fig. 6).

Circular dichroism spectra have also been carried out at basic and acid pHs (Fig. 7). The Cotton effect is totally suppressed at both pH = 3 and basic pHs.

Note that the model compound **p-THPP** did not exhibit any significant circular dichroism signal either in neat methanol or water : methanol, 9 : 1 (v/v).

Time-resolved absorption spectra

In general, porphyrins have intense triplet–triplet transient absorption, meaning both spectra and kinetics can be monitored by laser flash photolysis techniques. The triplet state of **CD-THPP** in neat methanol exhibits an absorption maximum at 450 nm, is photogenerated with a quantum yield $\Phi_T \sim 0.6$, decays in *ca.* 100 μs in nitrogen-flushed solutions and is quenched by oxygen with a diffusion-controlled rate constant. These properties are the same as those exhibited by **p-THPP** in the same solvent[37] and agree well with the similarity previously noted in the steady-state absorption and emission properties. However, when the laser flash photolysis measurements were carried out in water : methanol, 9 : 1 (v/v), the spectroscopic picture changed dramatically (Fig. 8).

The model compound **p-THPP** does not give any detectable signal in the 250–800 nm spectral range, whereas the absorption of the triplet state of **CD-THPP** is clearly monitored with a maximum at 460 nm.

The addition of **ADM** does not modify the shape of the triplet spectrum but influences significantly its top ΔA (A = absorbance), which decreases by *ca.* 50%. The trend observed reflects the behavior exhibited by the excited singlet state (see fluorescence results), the precursor of the triplet state, under the same experimental conditions. As far as the triplet decay of **CD-THPP** is concerned, it is interesting to note that it is slowed-down upon going from neat methanol to the water : methanol mixture (Fig. 9). The related first order rate constant decreases from *ca.* 1×10^5 s^{-1} to *ca.* 5×10^4 s^{-1}.

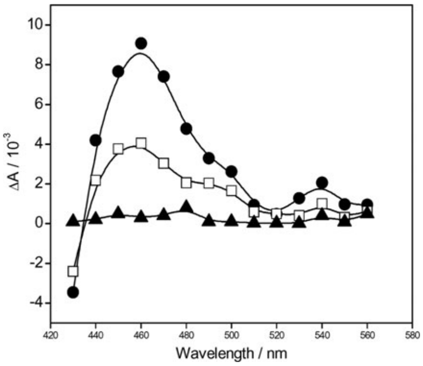


Fig. 8 Transient absorption spectra taken 1 μs after 532 nm laser excitation of N_2-saturated H_2O : methanol, 9 : 1 (v/v) optically matched solutions of **CD-THPP** (●), **CD-THPP** in the presence of **ADM** (□) and **p-THPP** (▲). $E_{532} \sim 3$ mJ.

Discussion

The combination of several spectroscopic techniques used in this work highlights the similarities and differences between the conjugate **CD-THPP** and model porphyrin **p-THPP** compounds, allowing insights to be gained into their molecular arrangements in two different solvents.

The CD receptor of **CD-THPP** plays a "silent" role in neat methanol. Indeed, the absorption, fluorescence and triplet–triplet spectra of the CD conjugate are virtually identical to those of the non-modified **p-THPP**.[37] Besides, the negligible dichroic band noted in MeOH, together with the absence of both intra- and intermolecular interactions between the porphyrin chromophore and the CD cavity emerging from the NMR data, suggest that, like the model **p-THPP**,[37] **CD-THPP** is present in a monomeric form in neat methanol.

Conversely, the role of the CD macrocycle is essential in differentiating the behavior of the conjugate with respect to the

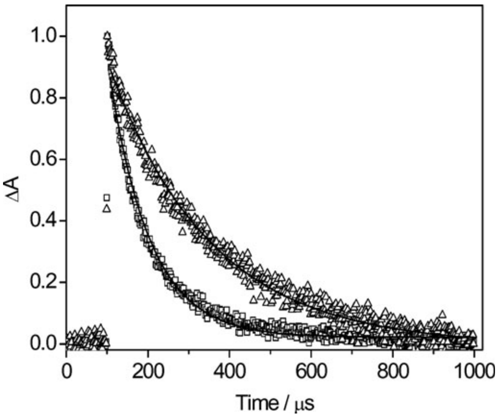


Fig. 9 Normalized decay traces with first order kinetic fits of **CD-THPP** in N_2-saturated methanol (□) and water : methanol, 9 : 1 (v/v) (△) solutions.

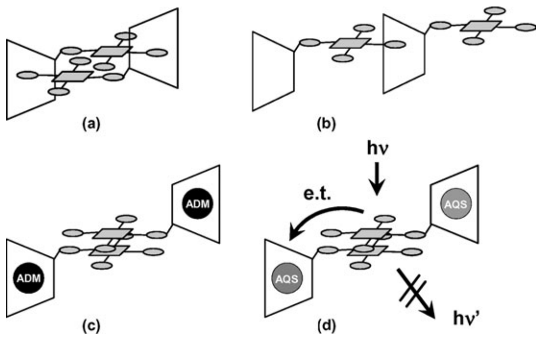
non-modified porphyrin in the mixture water : methanol, 9 : 1 (v/v). The strong hypochromism, accompanied by the broadening of the Soret band in the case of **p-THPP**, clearly suggest an effective aggregation under these experimental conditions. The effect of such an aggregation is drastically reflected in the photophysical properties of **p-THPP**. The fluorescence emission drops dramatically (see Fig. 4b) as a result of effective self-quenching processes and, accordingly, the population of the lowest triplet state of **p-THPP** is obviously precluded, as confirmed by the absence of any detectable transient signal in the laser flash photolysis experiments (Fig. 8). Finally, it is worth noting that, albeit weak, the emission yield of **p-THPP** exhibits a clear dependence on the excitation wavelength, pointing to the formation of many different types of aggregates characterized by different and low fluorescence quantum yields (inset Fig. 4b).[39]

This scenario significantly changes in the case of **CD-THPP**. Firstly, the Soret absorption of **CD-THPP** is narrower and more intense than that of the model compound, accounting for a reduction in aggregation phenomena. This is also confirmed by the response of **CD-THPP** to light stimuli. Indeed, we observe a significant revival of both the fluorescence emission and the typical triplet–triplet transient absorption. Moreover, the circular dichroism spectrum shows a strong split Cotton effect, consistent with an exciton coupling between the Soret transitions of two or more spatially-correlated chromophores.[40,41] This finding could account for the existence of an extremely stable dimeric aggregate of **CD-THPP** rather than a polymeric species, according to what proposed in the literature for ionic, water soluble porphyrins.[34,42] Structures (a) and (b) in the Scheme 1 depict two possible arrangements of **CD-THPP**, both of which account for the strong exciton coupling observed.

Of the two possible structures proposed for the supramolecular dimer, we believe that (a) as the more likely. The reasons supporting this proposal are now discussed. The independence of the $\Delta\varepsilon$ values of the circular dichroism spectra from the concentration of **CD-THPP** up to very low values cannot simply be ascribed in terms of a host–guest interaction driven by hydrophobic interactions, such as that proposed in case (b). Rather, a strong non-covalent interaction between the porphyrin rings has come into play. The absence of any relevant effect on the circular dichroism spectra, noted when free β-CD was added as a competitive host, is strictly in line with strong stacking interactions between aromatic rings stabilizing dimer formation. This pH independent behavior further corroborates the hypothesis of porphyrin–porphyrin interactions. In fact, the disappearance of the circular dichroism signal upon either protonation of the pyrrole nitrogens at acid pH or deprotonation of the hydroxyl groups at basic pH originates as result of electrostatic repulsion between the porphyrin rings. The photophysical measurements are also in good agreement with the model proposed. Indeed, the results obtained suggest the responses to light stimuli are mainly attributable to only one species, as confirmed by the almost complete independence of the fluorescence quantum yield from the excitation wavelength and the clean monoexponential decay of the triplet state of the porphyrin. In this context, it is worth noting that the triplet decay is slower than that observed in neat methanol (where **CD-THPP** is present as a monomer). The lengthening of the triplet decay is not uncommon with triplets upon incorporation into constrained media such as CDs, and is generally related to the protection exerted by the host cage against external quenching impurities and to a perturbation of the intersystem crossing to the ground state.[43] This finding is also of importance because it rules out a fast dissociation of the supramolecular structure in the excited triplet state.

The fact that the CD cavity plays a role in the arrangement of **CD-THPP** has been pointed out by using **ADM** as an effective competitive guest. The reduction in intensity of the dichroic band with increasing concentration of **ADM** (Fig. 6) reveals the disruption of the supramolecular structure. At first sight, the disappearance of the strong circular dichroism signal might suggest the formation of a monomeric species. However, this is not the case. Actually, the broadening of the Soret absorption and the decreasing of both the fluorescence emission and the triplet–triplet absorption, observed upon addition of **ADM**, clearly indicate that **CD-THPP** cannot be present as a monomer. In such a case, a behavior similar to that observed in neat methanol would have been reasonably expected. The model we propose is illustrated in Scheme 1(c). We believe that the incorporation of the guest within the CD cavity induces a new rearrangement of **CD-THPP**, probably in a sort of H-type dimer.[44] Such an aggregation mode would provide a satisfactory explanation for (i) the disappearance of the intense Cotton effect in the circular dichroism spectrum,[45] (ii) the broadening of the Soret band[16] and (iii) the reduced fluorescence emission as a result of the forbidden transition from the fluorescent S_1 state.[46]

The capability of the CD unit of **CD-THPP** to accommodate guest molecules able to communicate with the porphyrin chromophore under light stimuli is demonstrated by the fluorescence experiments carried out in the presence of **AQS**. This good electron acceptor is known to form a 1 : 1 complex with β-CD[36] and, similarly to other quinone derivatives, is expected to quench the excited singlet state of porphyrins by a photoinduced electron transfer mechanism.[47] Indeed, by assuming that the reduction potential of **CD-THPP** is similar to that of **p-THPP**, E^0(**CD-THPP**/**CD-THPP**$^{+\bullet}$), is *ca.* +1.1 V *vs.* SCE,[48] the reduction potential of **AQS**, E^0(AQS/AQS$^{\bullet}$), is *ca.* −0.6 V[49] and the energy of the excited singlet state of **CD-THPP**, $\Delta E_{0,0}$, is *ca.* 2.1 eV (obtained from the absorption and fluorescence spectra), it can be safely estimated that



Scheme 1 Schematic representation of potential arrangements for **CD-THPP**.

the thermodynamic balance based on the Rehm–Weller equation[50]

$$\Delta G = e[E^0(\text{CD-THPP/CD-THPP}^{+\bullet}) - E^0(\text{AQS/AQS}^{-\bullet})] - \Delta E_{0,0}$$

is negative, making the photoinduced process highly feasible. As shown in Fig. 5, the emission of **CD-THPP** is totally quenched by the addition of 5×10^{-4} M **AQS**. Since, at this low concentration, **AQS** cannot quench the porphyrin emission through a bimolecular, diffusion-controlled process, the fluorescence suppression observed is clearly attributable to an intramolecular photoinduced electron transfer *via* the CD cavity from the excited singlet state of the porphyrin to the included **AQS** (Scheme 1(d)). This hypothesis is further corroborated by the fact that addition of a competitive guest, such as **ADM**, capable of displacing **AQS** but unable to quench the porphyrin fluorescence by photoinduced electron transfer, leads to a partial recovery of the emission (Fig. 5), which becomes almost comparable to that obtained upon addition of **ADM** to **CD-THPP** alone (for comparison, see Fig. 4b).

Conclusions

The novel non-ionic conjugate **CD-THPP** exhibits a behavior virtually identical to the model non-modified **p-THPP** in methanol solution in terms of its spectroscopic properties and prevalence in a monomeric form. The scenario dramatically changes in water : methanol solution. While, in this solvent, the model compound **p-THPP** is non-responsive to light stimuli, as result of the formation of different types of aggregates, **CD-THPP** seems to self-associate prevalently into a sort of dimer, exhibiting a strong exciton splitting and a good response to photoexcitation. This aggregate is strongly stabilized by stacking interactions between the aromatic rings, in addition to the intermolecular hydrophobic porphyrin–cyclodextrin interactions. The incorporation of **ADM** within the CD cavity induces a new rearrangement of **CD-THPP**, probably in a sort of H-type dimer, characterized by negligible exciton splitting, reduced fluorescence emission and triplet–triplet absorption. This novel conjugate can also be exploited for the investigation of intramolecular photoinduced processes mediated by the CD cavity, as proven by the effective fluorescence quenching *via* photoinduced electron transfer, involving the excited porphyrin and a quinone accommodated within the CD cavity.

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References

1 (*a*) J. Szejtly, *Chem. Rev.*, 1998, **98**, 1743; (*b*) V. T. D'Souza and K. B. Lipkowitz, *Chem. Rev.*, 1998, **98**, 1741.

2 (*a*) R. P. Bonomo, S. Pedotti, G. Vecchio and E. Rizzarelli, *Inorg. Chem.*, 1996, **35**, 6873; (*b*) G. Maccarrone, E. Rizzarelli and G. Vecchio, *Polyhedron*, 2002, **21**, 1531; (*c*) V. Cucinotta, A. Giuffrida, D. La Mendola, G. Maccarrone, A. Puglisi, E. Rizzarelli and G. Vecchio, *J. Chromatogr., B: Biomed. Appl.*, 2004, **127**, 800.

3 (*a*) C. Donzé, E. Rizzarelli and G. Vecchio, *J. Inclusion Phenom.*, 1998, **31**, 27; (*b*) C. Donzé, E. Rizzarelli and G. Vecchio, *Supramol. Chem.*, 1998, **10**, 33; (*c*) H. Ikeda, M. Nakamura, N. Ise, N. Oguma, A. Nakamura, T. Ikeda, F. Toda and A. Ueno, *J. Am. Chem. Soc.*, 1996, **118**, 10980; (*d*) A. Ueno, A. Ikeda, H. Ikeda, T. Ikeda and F. Toda, *J. Org. Chem.*, 1999, **64**, 382; (*e*) Y. Liu, B.-H. Han, S.-X. Sun, T. Wada and Y. Inoue, *J. Org. Chem.*, 1999, **64**, 1487; (*f*) H. Ikeda, M. Nakamura, N. Ise, F. Toda and A. Ueno, *J. Org. Chem.*, 1997, **62**, 1411.

4 (*a*) S. Matsumura, S. Sakamoto, A. Ueno and H. Mihara, *Chem.–Eur. J.*, 2000, **6**, 1781; (*b*) M. A. Hossain, S. Matsumura, T. Kanai, K. Hamasaki, H. Mihara and A. Ueno, *J. Chem. Soc., Perkin Trans. 2*, 2000, 1527; (*c*) R. P. Bonomo, V. Bruno, E. Conte, G. De Guidi, D. La Mendola, G. Maccarrone, F. Nicoletti, E. Rizzarelli, S. Sortino and G. Vecchio, *Dalton Trans.*, 2003, 4406; (*d*) C. Pean, C. Creminon, A. Wijkhuisen, J. Grassi, P. Guenot, P. Jehan, J.-P. Dalbiez, B. Perly and F. Djedaini-Pilard, *J. Chem. Soc., Perkin Trans. 2*, 2000, 853; (*e*) E. Rizzarelli and G. Vecchio, *Coord. Chem. Rev.*, 1999, **188**, 343.

5 (*a*) Y. Liu, Y.-M. Zhang, S.-X. Sun, Y.-M. Li and R.-T. Chen, *J. Chem. Soc., Perkin Trans. 2*, 1997, 1609; (*b*) H. F. Nellissen, F. Venema, R. M. Uittenbogaard, M. C. Feiters and R. J. M. Nolte, *J. Chem. Soc., Perkin Trans. 2*, 1997, 2045; (*c*) A. Puglisi, G. Tabbì and G. Vecchio, *J. Inorg. Biochem.*, 2004, **98**, 969; (*d*) T. Kawabara, M. Takamura, A. Matsushita, H. Ikeda, A. Nakamura, A. Ueno and F. Toda, *J. Org. Chem.*, 1998, **63**, 8729.

6 R. Corradini, A. Dossena, G. Impellizzeri, G. Maccarrone, R. Marchelli, E. Rizzarelli, G. Sartor and G. Vecchio, *J. Am. Chem. Soc.*, 1994, **116**, 10267.

7 V. Thanabal and V. Krishnan, *J. Am. Chem. Soc.*, 1982, **104**, 3643.

8 K. Kano, R. Nishiyada, T. Asada and Y. Kuroda, *J. Am. Chem. Soc.*, 2002, **124**, 9937.

9 X. Gui, W. An, S. Shuang, F. Cheng and C. J. Dong, *J. Photochem. Photobiol., A*, 2005, **173**, 258.

10 P. Even and B. Boitrel, *Coord. Chem. Rev.*, 2006, **250**, 519.

11 A. A. Aksenova, Y. L. Sebyakin and A. F. Mironov, *Russ. J. Bioorg. Chem.*, 2003, **29**, 201.

12 J. J. Wu, H.-L. Ma, H.-S. Mao, Y. Wang and W.-J. Jim, *J. Photochem. Photobiol., A*, 2005, **173**, 296.

13 K. Sasaki, H. Nakagawa, X. Zhang, S. Sakurai, K. Kano and Y. Kuroda, *Chem. Commun.*, 2004, 408.

14 I. Hamachi, H. Takashima, Y.-Z. Hu, S. Shinkai and S. Oishi, *Chem. Commun.*, 2000, 1127.

15 I. Tabushi, *Coord. Chem. Rev.*, 1988, **86**, 1.

16 M. Kasha, *Radiat. Res.*, 1963, **20**, 55.

17 (*a*) R. V. Bensasson, E. J. Land, A. L. Moore, R. L. Crouch, G. Dirks, T. A. Moore and D. Gust, *Nature*, 1981, **290**, 329; (*b*) A. L. Moore, A. Joy, R. Tom, D. Gust, T. A. Moore, R. V. Bensasson and E. J. Land, *Science*, 1982, **216**, 982.

18 G. R. Geier and T. Sasaki, *Tetrahedron Lett.*, 1997, **38**, 3821.

19 N. Mamardashvili, I. Zh and O. I. Koifman, *Russ. J. Org. Chem.*, 2005, **41**, 787.

20 H. Iwamoto, Y. Yukimasa and Y. Fukazawa, *Tetrahedron Lett.*, 2002, **43**, 8181.

21 (*a*) K. Okamoto and S. Fukuzumi, *J. Am. Chem. Soc.*, 2003, **125**, 12416; (*b*) D. Gust, T. A. Moore, A. L. Moore, L. Leggett, S. Lin, J. M. De Graziano, R. M. Hermant, D. Nicodem, P. Craig, G. R. Seely and R. A. Nieman, *J. Phys. Chem.*, 1993, **97**, 7926.

22 (*a*) K. E. Splan, M. H. Keefe, A. M. Massari, K. A. Walters and J. T. Hupp, *Inorg. Chem.*, 2002, **41**, 619; (*b*) J. Andrèasson, J. Kajanus, J. Martensson and B. Albisson, *J. Am. Chem. Soc.*, 2000, **122**, 9844.

23 B. Jiang, S. Yang and W. W. Jones, Jr, *Chem. Mater.*, 1997, **9**, 2031.

24 (*a*) K. E. Splan, A. M. Massari and J. T. Hupp, *J. Phys. Chem. B*, 2004, **108**, 4111; (*b*) T. Hasobe, H. Imahori, H. Yamada, Y. Sato, K. Ohkubo and S. Fukuzumi, *Nano Lett.*, 2003, **3**, 409.

25 N. Kobayashi, U. Akida, K. Takatori, A. Ueno and T. Osa, *Heterocycles*, 1982, **19**, 11.

26 F. Venema, A. E. Rowan and R. J. M. Nolte, *J. Am. Chem. Soc.*, 1996, **118**, 257.

27 R. Breslow, X. Zhang and Y. Huang, *J. Am. Chem. Soc.*, 1997, **119**, 4535.

28 R. R. French, P. Holzer, M. G. Leunberger and W.-D. Woggon, *Angew. Chem., Int. Ed.*, 2000, **39**, 1267.

29 K. Lang, V. Kral, P. Kapusta, P. Kubát and P. Vašek, *Tetrahedron Lett.*, 2002, **43**, 4919.
30 Y. Kuroda, T. Hiroshige, T. Sera, Y. Shoroiwa, H. Tanaka and H. Ogoshi, *J. Am. Chem. Soc.*, 1989, **111**, 1912.
31 T. Kato and Y. Nakamura, *Heterocycles*, 1988, **27**, 4.
32 Y. Kuroda, M. Ito, T. Sera and H. Ogoshi, *J. Am. Chem. Soc.*, 1993, **115**, 7003.
33 K. Kano, R. Nishiyabu, T. Yamazaki and I. Yamazaki, *J. Am. Chem. Soc.*, 2003, **125**, 10625.
34 T. Carofiglio, R. Fornasier, V. Lucchini, L. Simonato and U. Tonellato, *J. Org. Chem.*, 2000, **65**, 9013.
35 R. P. Bonomo, V. Cucinotta, F. D'Alessandro, G. Impellizzeri, G. Maccarrone, G. Vecchio and E. Rizzarelli, *Inorg. Chem.*, 1991, **30**, 2708.
36 M. Rekharsky and Y. Inoue, *Chem. Rev.*, 1998, **98**, 1875.
37 R. Bonnett, D. J. McGarvey, A. Harriman, E. J. Land, T. G. Truscot and U.-J. Winfield, *Photochem. Photobiol.*, 1988, **48**, 271.
38 G. Pescitelli, S. Gabriel, Y. Wang, J. Fleischhauer, R. W. Woody and N. Berova, *J. Am. Chem. Soc.*, 2003, **125**, 7613.
39 R. F. Khairutdinov and N. Serpone, *J. Phys. Chem. B*, 1999, **103**, 761.
40 T. Nagata, A. Osuka and K. Maruyama, *J. Am. Chem. Soc.*, 1990, **112**, 3054.
41 O. Munro and H. M. Marques, *Inorg. Chem.*, 1996, **35**, 3768.
42 (*a*) K. Kano, M. Takei and S. Hashimoto, *J. Phys. Chem.*, 1990, **94**, 2181; (*b*) K. Kano, H. Minamizono, T. Kitae and S. Negi, *J. Phys. Chem. A*, 1997, **101**, 6118.
43 S. Monti and S. Sortino, *Chem. Soc. Rev.*, 2002, **31**, 287.
44 However, we cannot exclude the possibility that more than two units of **CD-THPP** can be staked in this new arrangement.
45 W.-W. Chen, J.-M. Yan, Y. Tagashira, M. Yamaguchi and K. Fujita, *Tetrahedron Lett.*, 1999, **40**, 891.
46 E. G. McRae and M. Kasha, *J. Chem. Phys.*, 1958, **28**, 271.
47 M. Z. Hoffman, F. Bolletta, L. Moggi and G. L. Hug, *J. Phys. Chem. Ref. Data*, 1989, **18**, 219.
48 K. Kalyanasundaram and M. Neunmann-Spallart, *J. Phys. Chem.*, 1982, **86**, 5163.
49 P. Wardman, *J. Phys. Chem. Ref. Data*, 1989, **18**, 1637.
50 D. Rehm and A. Weller, *Isr. J. Chem.*, 1970, **8**, 259.