

# Intramolecular excimer formation and sensing behavior of new fluorimetric probes and their interactions with metal cations and barbituric acids

C. Lodeiro<sup>a,\*</sup>, J.C. Lima<sup>a</sup>, A.J. Parola<sup>a</sup>, J.S. Seixas de Melo<sup>b,\*\*</sup>, J.L. Capelo<sup>a</sup>,  
B. Covelo<sup>a</sup>, A. Tamayo<sup>a</sup>, B. Pedras<sup>a</sup>

<sup>a</sup> REQUIMTE, Departamento de Química, Faculdade de Ciências e Tecnologia, Universidade Nova de Lisboa, 2829-516 Caparica, Portugal

<sup>b</sup> Departamento de Química, Universidade de Coimbra, 3004-535 Coimbra, Portugal

Received 17 June 2005; received in revised form 15 September 2005; accepted 19 September 2005

Available online 28 October 2005

## Abstract

A new family of compounds able to promote host–guest interactions with specific molecules (e.g., cyanuric and parabanic acids) and to coordinate metal ions, namely Zn(II) and Cu(II), has been synthesized and fully characterized. The new probes derive from the attachment of two methylaminopyrene units to the carbonyl precursor 2,6-bis(2-formylphenoxy)methylpyridine.

Its signalling properties result from the fluorescence emission properties, which reveal the existence of intramolecular excimer formation. The compounds have showed to be highly sensitive to the solvent and hydrogen ion concentration of the medium. Depending on these, different monomer-to-excimer fluorescence ratio is displayed by the two probes. The compound with a single pyrene unit revealed absence of excimer formation and was used as model compound. The overall results are discussed on the basis of the studied probes as potentially revealing molecular movements, off–on–off fluorescent photoinduced electron transfer (PET), host–guest interactions with specific compounds and of sensing metal ions.

© 2005 Elsevier B.V. All rights reserved.

**Keywords:** Barbituric acids; Excimers; Fluorescence; Receptor units; Supramolecular chemistry; Host–guest interactions

## 1. Introduction

The fluorimetric sensing of specific molecules by host–guest interactions is a research area of continuous development [1]. The sensitivity provided by the fluorimetric analysis (ranging from  $10^{-5}$  to  $10^{-8}$  M for the concentration of the fluorescent probe) provides a rich field for this development. Moreover, the additional circumstance of having a host (containing the fluorescent probe), whose fluorescence properties are not only based on the dependence of its fluorescence intensity but also presents new emission bands, resulting from specific movements and interactions with the guest molecules, provides additional and useful information on the type of host–guest interactions and on the effects (solvent, pH, temperature, etc.) affecting such interac-

tions. Therefore, the synthesis of new receptors with recognition ability towards a specific guest revealed a growing interest in the last decade. The synthesis of new ligand receptors of oxaza nature, capable of substrate recognition in solution, falls in this emerging field of selective sensing, due to its importance in basic and applied chemistry [2–5]. Among the existent fluorescent probes, pyrene reveals to be one of the most commonly used fluorophores due to its peculiar fluorescence properties: intense fluorescence signals, vibronic band dependence with the media [6], long lifetime values [7], and excimer formation [8–10]. Due to these peculiar properties and because of its chemical stability, it is also employed as a probe for solid-state studies [11,12], polymer association [13], polymer–polymer interactions, polymer–surfactant interactions, micelle critical concentration determinations, etc. [14–16].

Excimer-based sensors have often been used to probe host–guest interactions with anions and/or cations in organic solvent media [2,17–21]. Recent studies have extended pyrene emission and in particular, its ability for excimer formation to detect bases and discriminate [22] insertion in DNA and

\* Corresponding author. Tel.: +351 21 29 48 300/29 48 500x10923; fax: +351 21 29 48 385/29 48 550.

\*\* Corresponding author.

E-mail addresses: [lodeiro@dq.fct.unl.pt](mailto:lodeiro@dq.fct.unl.pt) (C. Lodeiro), [seixas@ci.uc.pt](mailto:seixas@ci.uc.pt) (J.S. Seixas de Melo).

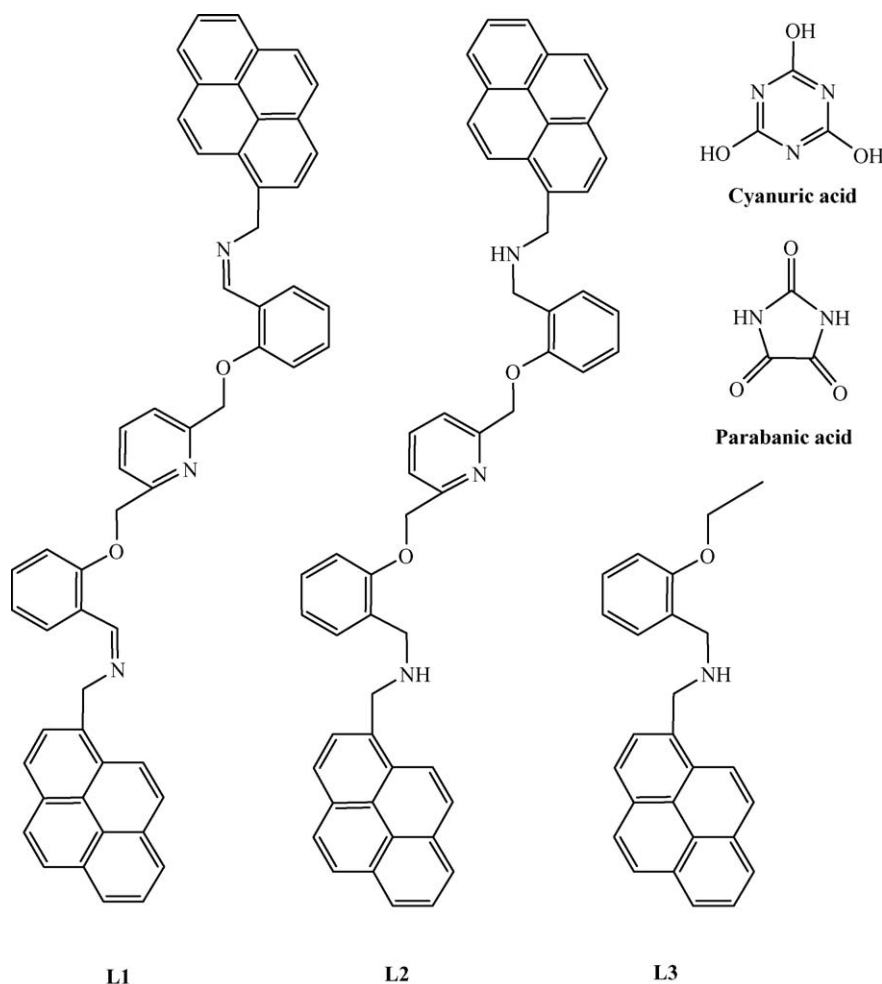


Fig. 1. Pyridine-pyrene ligands **L1** (pyren-1-ylmethyl-[2-(6-{2-[(pyren-1-ylmethylimino)-methyl]-phenoxy]methyl}-pyridin-2-ylmethoxy)-benzylidene]-amine), **L2** (pyren-1-ylmethyl-[2-(6-(2-[(pyren-1-ylmethyl)-amino]-methyl)-phenoxy]methyl)-pyridin-2-ylmethoxy]-benzyl]-amine) and **L3** ((2-ethoxy-benzyl)pyren-1-ylmethyl-amine). Cyanuric (2,4,6-trihydroxy-*s*-triazine) and parabanic acids (2,4,5-imidazolinetrione).

to selectively sense alkaline earth or transition metal cations [23–25].

Moreover, in supramolecular chemistry, the hydrogen bond is one of the most interesting non-covalent interactions favored by nature. Many researchers are interested in the hydrogen bond to investigate molecular recognition, self-assembly, and biomimicking [26–29].

The present work aims to describe the synthesis, chemical characterization and photophysical properties of two new pentadentate ligands containing one pyridine head linked to two benzenoid lateral units which are connected to two methylpyrene (fluorescent signaling probe) units (see Fig. 1). The ligands, here designated by **L1** and **L2**, possess a flexible structure consisting in two imine (**L1**) or two amine (**L2**) pyrenyl pendant-arms able to induce intramolecular excimer formation in solution, and to form hydrogen bonding interactions.

The main purpose of this study is to provide new basis for considering these probes for selective fluorescence sensing. We, therefore, describe the synthesis of metal complexes with **L1** (Schiff-base ligand) and the interaction of the two ligands (**L1** and **L2**) in solution with anions and/or cations. An additional

compound **L3**, consisting of a single pyrenyl unit, was synthesized for use as model compound in the photophysical studies.

## 2. Experimental

### 2.1. Materials and general methods

2,6-Bis(2-formylphenoxy)methylpyridine was prepared according to a published method [30]. 2,6-Pyridinedimethanol, salicylaldehyde, 2-ethoxybenzaldehyde, 1-pyrenemethylamine hydrochloride, triethylamine,  $\text{Zn}(\text{NO}_3)_2$ ,  $\text{Cu}(\text{NO}_3)_2$ ,  $\text{NaBH}_4$  were commercial products purchased from Aldrich Chemical Co. used without further purification. The solvents were of reagent grade chromasolv. Cyanuric (2,4,6-trihydroxy-*s*-triazine) and parabanic (2,4,5-imidazolinetrione) acids were purchased from Lancaster-Clariant and used without further purification.

Elemental analyses were performed by the University of Valencia, Spain and Universidade Nova de Lisboa (Monte de Caparica, Portugal).  $^1\text{H}$  NMR spectra were recorded using a Bruker WM-300. Positive-ion FAB mass spectra were recorded

by the University of Santiago de Compostela, Spain, on a Kratos MS50TC spectrometer using a 3-nitrobenzyl alcohol (MNBA) matrix.

The electronic absorption spectra were measured in the range 220–600 nm using a Shimadzu 2100 spectrophotometer and the fluorescence emission spectra on a Jobin Yvon-SPEX Fluorolog 3.22 spectrofluorometer. All solutions for the photophysical studies were not degassed, except for the time-resolved experiments. The concentration of ligands and metal ions were around  $10^{-6}$  or  $10^{-7}$  M in acetonitrile–water (99.5% water–0.5% acetonitrile), 100% acetonitrile or dichloromethane. In all photophysical studies, data were obtained with excitation at  $\lambda_{\text{exc}} = 316, 337$  or  $343$  nm and  $\lambda_{\text{em}} = 352, 370$  or  $520$  nm.

Fluorescence decays were measured using a home-built time-correlated single photon counting apparatus with an  $\text{N}_2$  filled IBH 5000 coaxial flashlamp as excitation source, Jobin-Ivon monochromator, Philips XP2020Q photomultiplier, and Canberra instruments time-to-amplitude converter and multichannel analyzer. Alternate measurements (1000 counts per cycle), controlled by Decay<sup>®</sup> software (Biodinâmica-Portugal), of the pulse profile at 337 or 356 nm and the sample emission were performed until  $1 \times 10^4$  to  $2 \times 10^4$  counts at the maximum were reached [31]. The fluorescence decays were analyzed using the modulating functions method of Striker with automatic correction for the photomultiplier “wavelength shift” [32].

$\text{Cu}^{2+}$  determinations were performed in a Varian model Zeeman spectrAA300 plus atomic absorption spectrometer in combination with an auto sampler; pyrolytic graphite-coated graphite tubes with platform were used [33]. For the determination of copper concentration, 20  $\mu\text{L}$  of solution were injected into the graphite furnace, where it was dried, ashed and atomized. The signal was measured in the peak area mode. Each

completed determination was followed by a 2 s clean-up cycle of the graphite furnace at  $2800^\circ\text{C}$ . During the drying, ashing, and clean-up cycles, the internal argon gas was passed through the graphite furnace at 300 mL/min. The internal argon gas flow was interrupted during the atomization cycle, but was restored for the clean-up cycle. The relative standard deviation among replicate was typically  $<5\%$ .

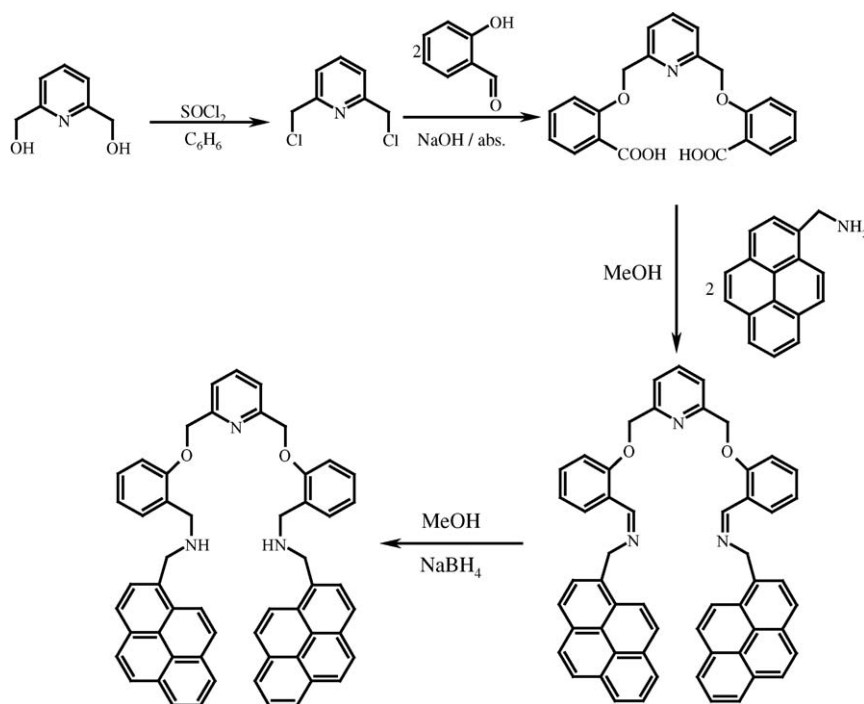
The FAAS measurements of  $\text{Zn}^{2+}$  were made using a Varian (Cambridge, UK) atomic absorption spectrometer model SpectrAA 20 plus equipped with a 10 cm burner head. A hollow-cathode lamp operated at 5 mA was used. The wavelength (nm) and slit width (nm) used were 213.9 and 1.0, respectively.

## 2.2. Synthesis

Reaction of 1-pyrenemethylamine hydrochloride and dicarbonyl compound 2,6-bis(2-formylphenoxy)methylpyridine gave a double Schiff-base that was either isolated as **L1** or reacted in situ with  $\text{NaBH}_4$  to obtain **L2** (see Scheme 1) as its chloridrate. The parent ligand **L3** was obtained by a similar procedure using as carbonyl 2-ethoxybenzaldehyde. Similar procedures have been used in the synthesis of macrocyclic systems derived from the same dicarbonyl precursor [34].

### 2.2.1. Synthesis of the Schiff-base ligand **L1**, pyren-1-ylmethyl-[2-(6-{2-[(pyren-1-ylmethylimino)-methyl]-phenoxy)methyl}-pyridin-2-ylmethoxy)-benzylidene]-amine

A solution of 1-pyrenemethylamine hydrochloride (2 mmol) in hot methanol (50 mL) in the presence of triethylamine (10 mmol) was added dropwise to a refluxing solution of 2,6-bis(2-formylphenoxy)methylpyridine (1 mmol) in the same hot solvent (50 mL). The resulting solution was gently refluxed with



Scheme 1. Scheme for the synthetic route of **L1** and **L2**.

magnetic stirring for ca. 4 h under Ar (g) atmosphere. The color of the solution changed to pale yellow, and a white precipitate formed upon cooling of the solution. It was filtered off, washed with cold absolute ethanol and diethyl ether and dried in vacuo. The ligand was recrystallized from chloroform-*n*-hexane as a pale cream solid.

**L1** Solid cream solvent; yield 49%; EA: found: C 82.80, H 5.05, N 5.25,  $C_{55}H_{41}N_3O_2 \cdot 1.25H_2O$  requires C 82.75; H 5.50; N 5.25%; IR (KBr disc):  $\nu(C=N)_{\text{imi}}$  1637  $\text{cm}^{-1}$ ,  $\nu(C=C)$  and  $\nu(C=N)$  1596–1488  $\text{cm}^{-1}$ ; mass spectrum (positive-ion FAB):  $m/z$  775 [**L1** + H]<sup>+</sup>; <sup>1</sup>H NMR (CD<sub>3</sub>CN),  $\delta$ (ppm): 8.95 (s), 2H (imine), 8,5 (d), 2H (Py); 8,2(t), 1H (Py); 7,5–6.0(m) 26H (pyrene- $C_6H_6$ ); 5,6(s), 4H (pyrene- $CH_2$ ); 5.1(s), 4H (Py- $CH_2$ ). The ligand is air stable, soluble in acetonitrile, chloroform, dichloromethane, 95% water–5% acetonitrile and insoluble in absolute ethanol and diethyl ether.

### 2.2.2. Synthesis of the reduced ligand **L2** (pyren-1-ylmethyl- $\{2-[6-(2-\{[(\text{pyren-1-ylmethyl-amino})\text{-methyl}]\text{-phenoxyethyl})\text{-pyridin-2-ylmethoxy}]\text{-benzyl}\}$ -amine

A solution of 1-pyrenemethylamine hydrochloride (2 mmol) in hot methanol (50 mL) in the presence of triethylamine (10 mmol) was added dropwise to a refluxing solution of 2,6-bis(2-formylphenoxyethyl)pyridine (1 mmol) in the same hot solvent (50 mL) during 1 h. The resulting solution was gently refluxed with magnetic stirring for ca. 2 h under Ar (g) atmosphere. The color of the solution changed to yellow. The reaction mixture was cooled over an ice-bath, and then NaBH<sub>4</sub> (20 mmol) was added carefully in solid incremental amounts with stirring. After effervescence had stopped, the solution was filtered, and was reduced to dryness by rotary evaporation. The obtained oil was dissolved in water, and three different extractions with chloroform (150 mL) were made. The organic phase was dried over anhydrous magnesium sulphate. The solvent was removed by rotary evaporation to ca. 10–15 mL and diethyl ether was added producing an oil product that was decanted and dried in vacuo. The oil was redissolved in absolute ethanol and 10 M HCl was address dropwise, leading to the formation of a pale yellow precipitate that was filtered off, washed with cold methanol and diethyl ether and dried in vacuo.

**L2** Pale yellow solid; yield 40%; EA: found: C 67.10, H 5.20, N 4.45,  $C_{55}H_{45}N_3O_2 \cdot 3HCl \cdot 5H_2O$  requires C 67.45, H 5.95, N 4.30%; IR (KBr disc):  $\nu(NH)$  3200  $\text{cm}^{-1}$ ,  $\nu(C=C)$  and  $\nu(C=N)$  1597, 1453  $\text{cm}^{-1}$ . Mass spectrum (positive-ion FAB):  $m/z$  780 [**L2** + H]<sup>+</sup>; <sup>1</sup>H NMR (CD<sub>3</sub>CN),  $\delta$ (ppm): 8,3 (d), 2H (Py); 8,2(t), 1H (Py); 8,0–6.4(m) 26H (pyrene- $CH_6$ ); 4,8(s), 4H (pyrene- $CH_2$ ); 4.4(s), 4H (Py- $CH_2$ ); 4.3(s), 4H (C<sub>6</sub>H<sub>6</sub>- $CH_2$ ). The ligand is air stable. Soluble in acetonitrile, mixtures of acetonitrile–water, moderately soluble in dichloromethane, chloroform and water, and insoluble in diethyl ether.

### 2.2.3. Synthesis of the reduced parent ligand **L3** (2-ethoxy-benzyl)pyren-1-ylmethyl-amine

A solution of 1-pyrenemethylamine hydrochloride (0.5 mmol) in hot methanol (25 mL) in the presence of triethylamine (5 mmol) was added dropwise to a refluxing solution of 2-ethoxybenzaldehyde (0.5 mmol) in the same

hot solvent (25 mL) during 1 h. The resulting solution was gently refluxed with magnetic stirring for ca. 2 h under Ar (g) atmosphere. The color of the solution changed to yellow. The reaction mixture was cooled over an ice-bath, and then solid NaBH<sub>4</sub> (10 mmol) was added carefully in solid incremental amounts with. After effervescence had stopped, the solution was filtered, and was reduced to dryness by rotary evaporation. The oil obtained was dissolved in water, and chloroform (100 mL) was added in three times. The organic phase was dried over anhydrous magnesium sulphate. The solvent was removed by rotary evaporation to ca. 10–15 mL and diethyl ether was added producing an oil product, was decanted and dried in vacuo. The oil was redissolved in absolute ethanol and 10 M HCl was address dropwise, leading to the appearance of a pale yellow precipitate, that was filtered off, washed with cold methanol and diethyl ether and dried in vacuo.

**L3** Pale yellow solid; yield 50%; EA: found: C 77.25, H 6.05, N 3.45,  $C_{26}H_{23}NO \cdot HCl$  requires C 77.70, H 6.00, N 3.50%; IR (KBr disc):  $\nu(NH)$  3220  $\text{cm}^{-1}$ ,  $\nu(C=C)$  and  $\nu(C=N)$  1595, 1450  $\text{cm}^{-1}$ ; mass spectrum (positive-ion FAB):  $m/z$  367 [**L3** + H]<sup>+</sup>. The ligand is air stable. Soluble in acetonitrile, acetonitrile–water, moderately soluble in dichloromethane, chloroform and water, and insoluble in diethyl ether.

## 2.3. Synthesis of the metal complexes with the Schiff-base ligand **L1**

### 2.3.1. General procedure for $[M_2L1][NO_3]_4$

A solution of 1-pyrenemethylamine hydrochloride (2 mmol) in hot methanol (25 mL) in the presence of triethylamine (10 mmol) was added dropwise to a refluxing solution of 2,6-bis(2-formylphenoxyethyl)pyridine (1 mmol) in the same hot solvent (25 mL). The resulting solution was gently refluxed with magnetic stirring for ca. 1 h under Ar (g) atmosphere.  $M(NO_3)_2 \cdot xH_2O$  (1 mmol) ( $M = Zn$  or  $Cu$ ) was dissolved in hot methanol (15 mL) and was added dropwise, with magnetic stirring and heating of the solution was maintained during the addition. The solution was refluxed for 2 h and then allowed to cool to room temperature overnight with stirring. Both complexes precipitated directly, were filtered off, washed with cold absolute ethanol then diethyl ether and dried in vacuo. The complexes are air stable, soluble in acetone, dimethylformamide, methanol, acetonitrile and dichloromethane, moderately soluble in chloroform, and insoluble in water and absolute ethanol.

**L1Cu<sub>2</sub>** Dark green solid; yield 56%; EA: found: C 55.45, H 3.60, 8.30, Cu 10.60,  $C_{55}H_{41}N_3O_2Cu_2(NO_3)_4 \cdot 2H_2O$  requires C 55.65, H 3.80, N 8.25, Cu 10.70%; IR (KBr disc):  $\nu(C=N)_{\text{imi}}$  1648  $\text{cm}^{-1}$ ,  $\nu(C=C)$  1598, 1450  $\text{cm}^{-1}$ ,  $\nu(NO_3)$  1384, 1303, 838, 814  $\text{cm}^{-1}$ ; mass spectrum (positive-ion FAB):  $m/z$  775 [**L1** + H]<sup>+</sup> (100%);  $m/z$  839 [**L1** + Cu]<sup>+</sup>;  $m/z$  904 [**L1** + Cu<sub>2</sub>]<sup>+</sup>. The complex is air stable, soluble in acetonitrile, absolute ethanol, methanol, chloroform and dichloromethane. In solution, it slowly decomposes.

**L1Zn<sub>2</sub>** Yellow solid; yield 81%; EA: found: C 56.30, H 3.85, N 8.15, Zn 11.20,  $C_{55}H_{41}N_3O_5Zn_2(NO_3)_4 \cdot H_2O$  requires C 56.30, H 3.90, N 8.35, Zn 11.15%; IR (KBr disc):  $\nu(C=N)_{\text{imi}}$  1646  $\text{cm}^{-1}$ ,  $\nu(C=C)$  1598, 1455  $\text{cm}^{-1}$ ,  $\nu(NO_3)$  1384, 1287,

826, 752  $\text{cm}^{-1}$ ; mass spectrum (positive-ion FAB):  $m/z$  775  $[\mathbf{L1} + \text{H}]^+$  (100%);  $m/z$  840  $[\mathbf{L1} + \text{Zn}]^+$ ;  $m/z$  968  $[\mathbf{L1Zn}_2(\text{NO}_3)]^+$ . The complex is air stable, soluble in acetonitrile, absolute ethanol, dichloromethane, and insoluble in diethyl ether and petroleum ether.

### 3. Results and discussion

#### 3.1. Synthesis of the free ligands

Yields, microanalytical and spectroscopic data for the ligands are presented in Section 2. Direct reaction between 2,6-bis(2-formylphenoxy)methylpyridine and two equivalents of 1-pyrenemethylamine hydrochloride in methanol gives the corresponding [1+2] Schiff-base podand **L1** (Scheme 1). In previous papers, we have already reported the use of this dicarbonylic precursor in the synthesis of different macrocyclic ligands, in the absence of templating agents [34]. Treatment of both imine bonds with  $\text{NaBH}_4$  gives the corresponding reduced diamine system, **L2**, as the major product. The corresponding reduced monofluorophoric compound **L3**, synthesized to obtain a parent compound for spectroscopic studies, was prepared by an in situ reduction by  $\text{NaBH}_4$  of the corresponding Schiff-base, result of the condensation of 1-pyrenemethylamine hydrochloride with 2-ethoxybenzaldehyde. **L1**, **L2** and **L3** were isolated as solids in 49, 40 and 50% yields, respectively. The FAB mass spectra show peaks corresponding to  $[\mathbf{L1} + \text{H}]^+$ , at 775  $m/z$ ;  $[\mathbf{L2} + \text{H}]^+$ , at 780  $m/z$  and  $[\mathbf{L3} + \text{H}]^+$ , at 367  $m/z$ . The IR spectrum of **L1** presents a band at ca. 1637  $\text{cm}^{-1}$  assignable to the imine groups, as well as bands at ca. 1596 and 1448  $\text{cm}^{-1}$ , related to  $\nu(\text{C}=\text{C})$  and  $\nu(\text{C}=\text{N})_{\text{py}}$ . For **L2** and **L3**, the band at ca. 3200  $\text{cm}^{-1}$  confirms the presence of the amine groups. The  $^1\text{H}$  NMR spectrum of **L1** in  $\text{CD}_3\text{CN}$  shows a peak at 8.95 ppm, indicating the presence of the imine protons; no signals corresponding to the formyl or amine protons are present.

#### 3.2. Synthesis of metal complexes with **L1**

The reactions between 2,6-bis(2-formylphenoxy)methylpyridine with two equivalents of 1-pyrenemethylamine hydrochloride and hydrated metal nitrates in methanol under the conditions described in Section 2 gave analytically pure products  $[\text{M}_2\mathbf{L1}](\text{NO}_3)_4 \cdot x\text{H}_2\text{O}$  ( $\text{M} = \text{Zn}(\text{II})$  and  $\text{Cu}(\text{II})$ ). The analytical data for the complexes are presented in Section 2.

The FAB mass spectra feature in all cases a peak at  $m/z$  775 ( $[\mathbf{L1} + \text{H}]^+$ ) as the most intense one, confirming the stability of the ligand in the metal complexes. Peaks assignable to the fragments  $[\text{Cu}\mathbf{L1}]^+$ ,  $[\text{Cu}_2\mathbf{L1}]^+$ ,  $[\text{Zn}\mathbf{L1}]^+$  and  $[\text{Zn}_2\mathbf{L1}(\text{NO}_3)]^+$  can be observed for the dinuclear complexes of **L1**.

The IR spectra confirm the existence of the ligand in the complexes, with the presence of absorption bands due to imine groups at ca. 1648–1646  $\text{cm}^{-1}$  and the absence of bands due to  $\nu(\text{C}=\text{O})$  or  $\nu(\text{NH}_2)$  [35]. All spectra exhibit medium to strong bands at ca. 1598 and 1455  $\text{cm}^{-1}$  as expected for the two highest energy pyridine-ring vibrations [36]. The shift of the imine and pyridine bands to higher wavenumbers in the complexes,

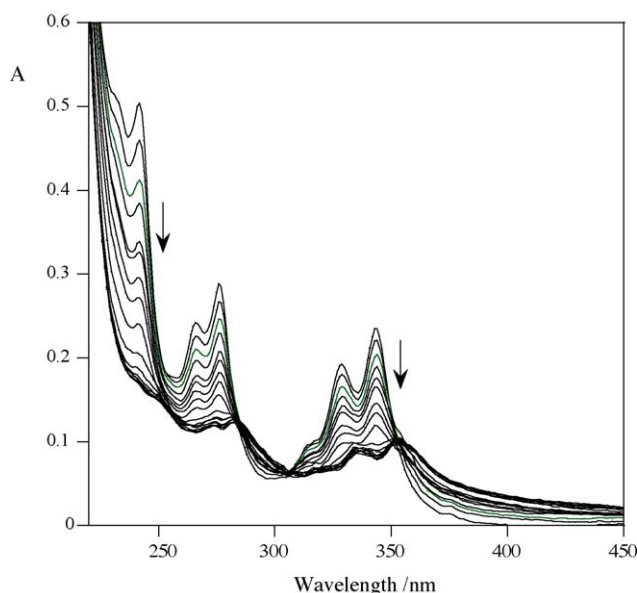


Fig. 2. Absorption spectra of ligand **L2** in 95.5% water–0.5% acetonitrile solution as function of pH;  $\lambda_{\text{exc}} = 340 \text{ nm}$ ,  $T = 298 \text{ K}$ ,  $[\mathbf{L2}] = 6.00\text{E}^{-6} \text{ M}$ ; pH 1.32, 1.69, 1.97, 2.05, 2.95, 2.73, 3.84, 4.45, 5.5, 6.35, 6.78, 7.59, 5.60, 9.0, 9.60, 10.65 and 11.10.

compared to those of the free ligands, may be indicative of coordination via the imine and pyridine nitrogen atoms [37,38]. Moreover, the broad absorption band in the region  $3450 \text{ cm}^{-1}$  present in both complexes is probably due to the existence of lattice and/or co-ordinated water in the molecules [39].

Attempts to obtain the corresponding mononuclear complexes with **L1** were unsuccessful.

#### 3.3. Spectroscopic behavior for the free ligands

Fig. 2 shows the absorption spectra of **L2** as a function of pH in water:acetonitrile 99.5:0.5 (v/v) solution. Pure water could not be used due to the low solubility of the ligand in this solvent. At acidic pH values, the absorption spectra resembles that of pyrene, but with the increase in the pH value of the medium, a shift towards longer wavelengths and a progressive broadening is observed. The origin of this new band, at alkaline pH values, probably results from intramolecular ground-state association between the two pyrene units of the compound.

The emission spectra of **L2**, obtained at several pH values is presented in Fig. 3. The total fluorescence emission intensity is largely dependent on the pH of the medium and consequently on the protonation state of the compound. This arises as a consequence of the deprotonation of the amino group, which induces the quenching of the excited fluorophore by electron transfer [40].

As expected from a common and general behavior found for similar compounds (amino receptors possessing a variety of fluorophores), the more protonated form was expected to be the most emissive species (see Fig. 3) since electron transfer, from protonated amines to the excited fluorophores, is not thermodynamically allowed [40]. However, in our systems, the presence of a pyridine unit changes this behavior, since

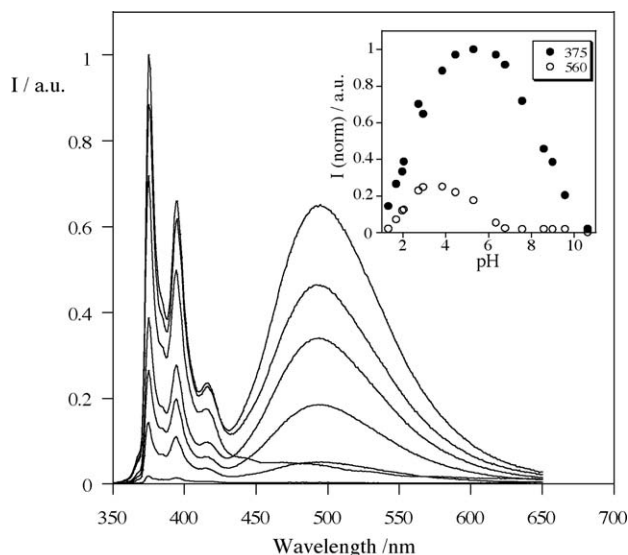


Fig. 3. Emission spectra of ligand **L2** in 95.5% water–0.5% acetonitrile solution as function of pH;  $\lambda_{\text{exc}} = 340$  nm,  $T = 298$  K,  $[\text{L2}] = 6.00 \times 10^{-6}$  M; pH 1.32, 1.69, 2.04, 3.84, 5.3, 7.6 and 11.10.

protonated pyridine quenches the excited pyrene fluorophore. This was previously described by de Silva et al. [41] for a system provided with an anthracene fluorophore linked to a pyridine-ring. This photoactive supramolecular system shows quenching due to photoinduced electron transfer (PET) in two situations: (i) at low pH values, when the pyridine unit is protonated and both amino groups are also protonated (“reverse logic on–off switching”) and (ii) at neutral–basic pH values, when the pyridine unit is unprotonated and at least one amino group is unprotonated. Similar to the work reported by de Silva et al. [41], it is possible to construct with this sensor, a pH window with an “off–on–off” behavior between pH 1–10 based on the monomer emission and between pH 1 and 6 when the excimer emission is taken into account (see Figs. 3 and 4).

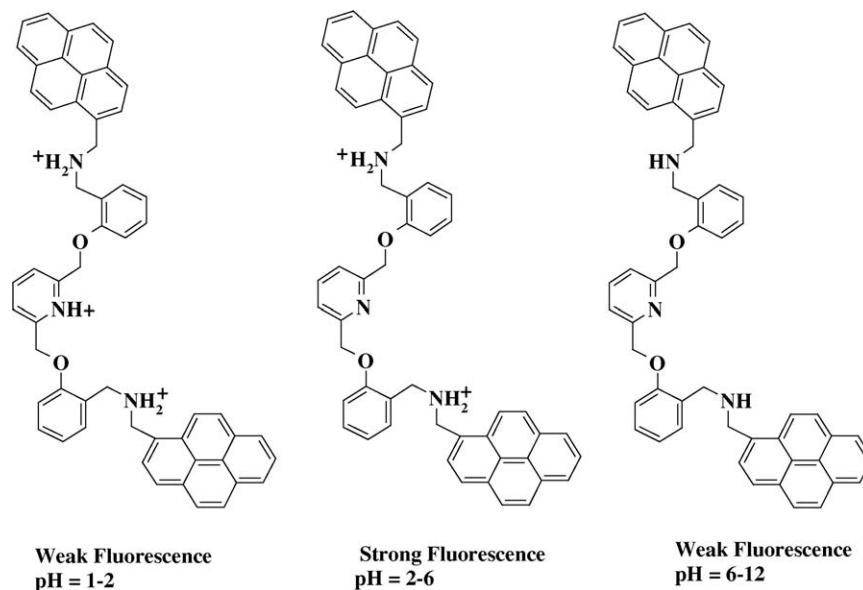


Fig. 4. Schematic representation of the operation of a fluorescent PET sensor for a pH window.

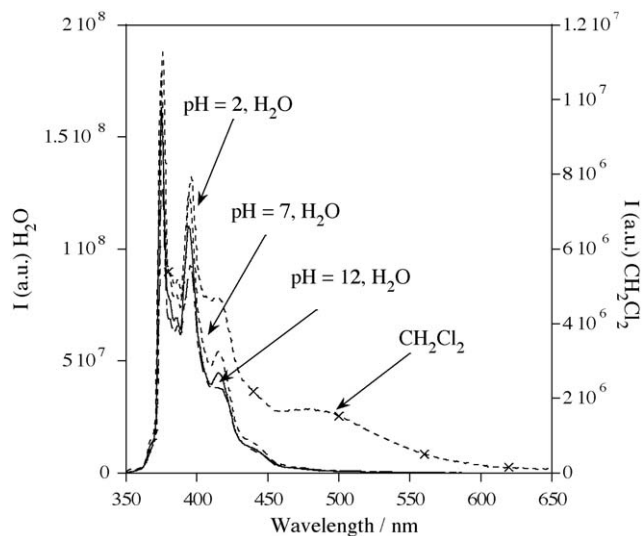


Fig. 5. (A) Emission spectra of the **L1** in water–acetonitrile 95.5% water–0.5% acetonitrile solution as function of pH;  $\lambda_{\text{exc}} = 340$  nm,  $[\text{L1}] = 8.85 \times 10^{-6}$  M. (B) Emission spectra of the Schiff-base ligand **L1** in water–acetonitrile (95.5% water–0.5% acetonitrile) and in dichloromethane solutions ( $\lambda_{\text{exc}} = 340$  nm,  $[\text{L1}] = 6.85 \times 10^{-6}$  M).

The most interesting feature of the emission spectra of the present systems, exemplified in Fig. 5 for **L1**, is the appearance of a red-shifted, non-structured band, which is known as the fingerprint of an excimer emission. This structureless band does not occur with the compounds containing one single terminal pyrene unit (compound **L3**) and is absent in water in the case of **L1**, probably due to partial hydrolysis of the C=N bonds present in this ligand. Aminomethylpyrene emission itself does not show this red-shifted emission band, which excludes the possibility that this emission arises from an emissive charge transfer state involving the deprotonated amine and the pyrene unit. Besides, the observation that an increase in pH results in the decrease of the fluorescence intensity, it is also worth

noting that the excimer emission contribution (relative to the monomer emission) decreases faster than the monomer emission. In fact, from the inset in Fig. 3, a sharp decrease of the fluorescence intensity of **L2** excimer's band, starting at  $\text{pH} \cong 4$ , can be observed, whereas this decrease in the monomer band only arises for  $\text{pH} \approx 6$ , i.e., at a higher pH value.

### 3.4. Time-resolved data for **L1** and **L2**

The fluorescence decays obtained at pH 10.7 revealed the appearance of an additional short-lived component of approximately 4.9 ns that increases in importance when going from the monomer to the excimer emission. Moreover this component begins to be present at pH 5.7, with the concomitant disappearance of the rising component. The existence of dimer structures in pyrene- or naphthalene-containing molecules has been recently equated as influencing the vibronic intensities in the monomer band as well as giving rise to short-lived components [13,42].

A deeper understanding of the occurring phenomena in the excited state of the studied compounds demands the knowledge of its dynamic behavior. In non-protic solvents, such as dichloromethane, the **L1** excimer emission is present (Fig. 5). In fact, the fluorescence decays obtained in dichloromethane solution revealed the existence of three components; however, only two of the components are coupled to each other and the longer lifetime component, with a very low weight at 520 nm, could be attributable to free methylaminopyrene resulting from some degree of hydrolysis [43]. The obtained decay times and associated pre-exponential factors for **L1** and **L2** are summarized in Table 1.

The observation of data in Table 1 shows the simultaneous static and dynamic routes for excimer formation. This is attested by the presence of the negative pre-exponential associated with the rise-time (dynamic excimer formation) and by the fact that the sum of the pre-exponential factors at the excimer emission wavelength is not 0 (static contribution). Also of relevance in Table 1, and as previously discussed, is to observe a longer lifetime component that should be attributable to partial hydrolysis of **L1**, giving rise to free protonated methylaminopyrene whose fluorescence maximum is mainly found at 370 nm ( $A_3$  pre-exponential at 370 nm equal to 0.41 in Table 1).

With the previous observation that compound **L1** probably undergoes partial hydrolysis in water, fluorescence decays studies in this solvent were performed only with ligand **L2**. The fluorescence decays of ligand **L2** in water at pH 3 (where excimer emission is more intense) revealed to be best fitted with a sum of two exponentials both at the monomer and excimer emission regions (see Table 1). Although a third exponential (long component) was generally needed to properly fit the decays at the monomer emission, this was found to be related to the presence of a very small amount (less than 0.8% of the total emission) of the pirenymethylamine chloride (see Section 2) starting material in the synthesis of **L1** and **L2**. The sum of pre-exponential factors at the excimer emission wavelength is never equal to 0. If excimer formation would occur only through diffusional encounter between one excited pyrene and one non-excited pyrene, this pre-exponential sum would amount to 0, thus revealing that no fraction of pre-associated pyrenes was present. However, this is not the case and some degree of ground-state association must be considered.

### 3.5. Spectroscopic studies of the interaction of **L2** with $\text{Zn}^{2+}$ , $\text{Cu}^{2+}$ , parabanic and cyanuric acids

The formation of complexes with  $\text{Zn}^{2+}$ ,  $\text{Cu}^{2+}$ , parabanic and cyanuric acids was studied in aqueous solution at different molar ratios.

### 3.6. $\text{Zn}^{2+}$ and $\text{Cu}^{2+}$ complexes

Addition of two equivalents of Cu(II) or Zn(II) to **L1** in dichloromethane leads to the disappearance of the excimer band observed in the free system (see Fig. 6, left). This behavior suggests that upon complexation, the two pyrene units are not able to enter in close proximity.

Fig. 6 (right) shows the normalized (at the monomer emission maxima) fluorescence spectra of a solution of **L2** in 95.5% water–0.5% acetonitrile solution and in the presence of two equivalents of Cu(II) and Zn(II). Upon addition of the metal ion in the proportion **L2**:metal 1:2, the excimer fluorescence decreases relative to free **L2** ligand. This could partially be explained if the formation of the **L2**:M complexes establishes a conformation where the two pyrene-rings will be, in the ground-state, at a less favorable inter-chromophoric bond distance.

Table 1  
Decay times and associated pre-exponential factors for **L1** in dichloromethane and **L2** in water at different pH values ( $T=293\text{ K}$ )

Compound	Solvent	$\lambda_{\text{em}}$ (nm)	$\tau_1$ (ns)	$\tau_2$ (ns)	$\tau_3$ (ns)	$A_1$	$A_2$	$A_3$	$\chi^2$
<b>L1</b>	$\text{CH}_2\text{CL}_2$	370	3.1	43.9	128.4	0.54	0.05	0.41	0.98
		520				−0.55	0.97	0.03	1.12
<b>L2</b>	Water pH 1.2	390	3.3	11.2	135.2	0.73	0.16	0.11	1.01
		560				−0.27	1.00	–	1.00
	Water pH 3.0	390	7.7	37.3	102.5	0.56	0.24	0.20	1.16
		560				−0.67	1.00	–	1.14
	Water pH 5.5	390	9.8	42.0	109.9	0.23	0.34	0.43	1.07
		560				−0.20	1.00	–	1.07

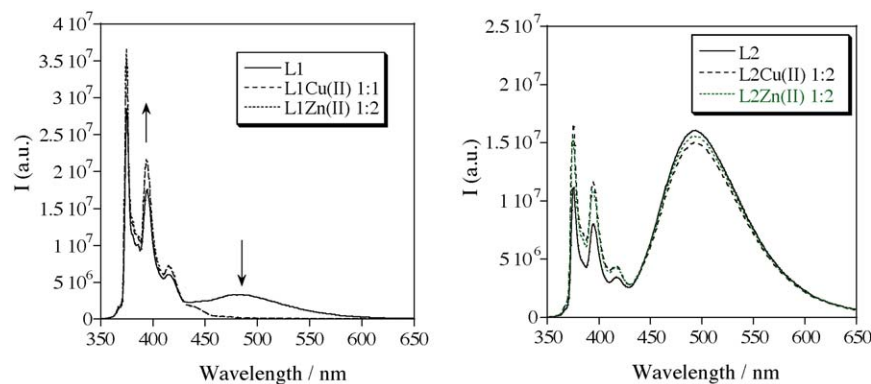


Fig. 6. Emission spectra of the ligand **L1** in  $\text{CH}_2\text{Cl}_2$  (black line) (left) **L2** in 95.5% water–0.5% acetonitrile solution (black line) (right) and in the presence of two equivalents of Cu(II) and Zn(II) ( $\lambda_{\text{exc}} = 343 \text{ nm}$ ,  $25^\circ\text{C}$ ,  $[\text{L1}, \text{L2}] = 6.00\text{E}^{-6} \text{ M}$ ) at pH 5.15.

### 3.7. Cyanuric acid (CnAc)

Fig. 7 shows the emission spectra of ligand **L2** with increasing concentrations of cyanuric acid (CnAc). Addition of CnAc leads to a decrease in the emission intensity of the excimer band with a concomitant increase in the intensity of the monomer band; this is very clear in the inset of Fig. 7, where the excimer-to-monomer ratio is plotted.

Fitting the emission intensities at both the excimer and monomer emission maxima allows to calculate the 1:1 association constant between **L2** and CnAc as  $\log K = 5.13 \pm 0.06$ . This value is corroborated by using Specfit/32<sup>TM</sup> v. 3.0, which leads to  $\log K = 5.21 \pm 0.06$ . These values are, however, uncertain since plateaus are not attained due to precipitation of the host–guest supermolecule. In fact, the spectrum of **L2**·CnAc calculated by Specfit has still excimer emission showing that the ligand is not totally associated with CnAc (although the possibility that the supermolecule is still flexible enough to form excimer could not be excluded).

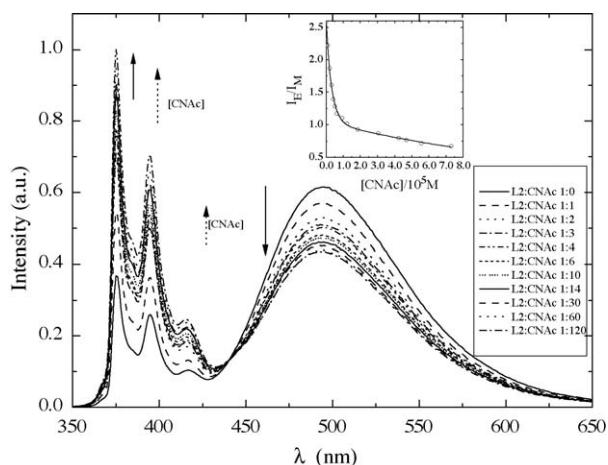


Fig. 7. Fluorescence emission spectra dependence of the **L2** in water–acetonitrile (95.5% water–0.5% acetonitrile) mixture;  $\lambda_{\text{exc}} = 340 \text{ nm}$ ,  $[\text{L2}] = 8.85\text{E}^{-6} \text{ M}$  in the presence of increased amounts of cyanuric acid (CnAc). Shown as inset is the plot of the excimer-to-monomer intensity ratio as a function of the molar concentration of CnAc;  $T = 293 \text{ K}$  and  $K$  (ca.) =  $1.10\text{E}^{-5}$ .

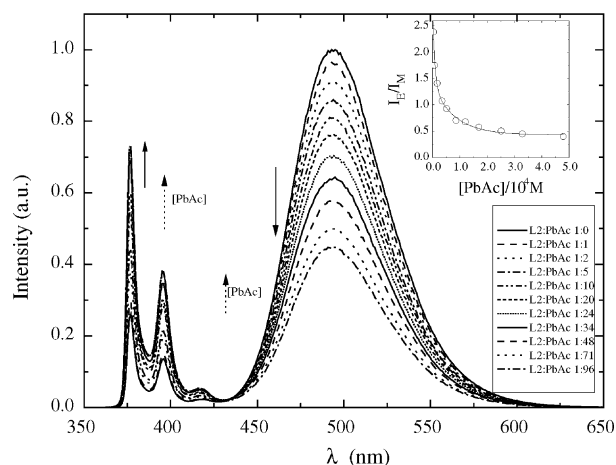


Fig. 8. Fluorescence emission spectra dependence of the **L2** in water–acetonitrile (95.5% water–0.5% acetonitrile) mixture;  $\lambda_{\text{exc}} = 340 \text{ nm}$ ,  $[\text{L2}] = 8.85\text{E}^{-6} \text{ M}$  in the presence of increased amounts of parabanic acid (PbAc). Shown as inset is the plot of the excimer-to-monomer intensity ratio as a function of the molar concentration of PbAc;  $T = 293 \text{ K}$  and  $K$  (ca.) =  $1.10\text{E}^{-5}$ .

### 3.8. Parabanic acid (PbAc)

The fluorescence emission spectra of ligand **L2** upon increasing concentrations of parabanic acid is shown in Fig. 8. Like in the case of the cyanuric acid complex, it is clear that a decrease in the excimer-to-monomer ratio occurs with the addition of increasing amounts of PbAc. In this case, however, simultaneous fitting of emission intensities at both monomer and excimer emission maxima is hardly achievable by hand. Specfit/32<sup>TM</sup> is able to fit the data with a 1:1 **L2**:PbAc species but the best fit clearly emerges when a supramolecular species **L2**:PbAc 1:2 is included in the model, giving  $\log K_1 = 4.6 \pm 0.1$  and  $\log K_2 = 3.2 \pm 0.1$ . In this case, the spectrum associated to **L2**·(PbAc)<sub>2</sub> shows only monomer emission, suggesting that two guest molecules are necessary to prevent excimer formation [44].

## 4. Conclusions

Two bischromophoric sensors, and one monochromophoric system based on pyrene emission have been synthesized and



fully characterized. Their emission properties were studied in water–acetonitrile solutions as a function of pH and in the presence of Zn(II), Cu(II) and barbituric acids, such as cyanuric and parabanic acids. The isolated ligand **L2** behaves in fluorescence as a off–on–off system both in monomer and in excimer emissions. While the addition of metal ions does not significantly quench the emission, the barbituric acids strongly reduce excimer formation.

## Acknowledgments

This work has been supported by the Fundação para a Ciência e a Tecnologia (Project POCI/QUI/55519/2004 FCT-FEDER) (Portugal). BC thanks Caixanova (Spain) by a post-doctoral grant, AT thanks Departament d'Universitats Recerca i Societat de la Informació of the Catalan Government (Spain) by the Grant FI2002-00320.

## References

- [1] A.P. deSilva, H.Q.N. Gunaratne, T. Gunnlaugsson, A.J.M. Huxley, C.P. McCoy, J.T. Rademacher, T.E. Rice, Signaling recognition events with fluorescent sensors and switches, *Chem. Rev.* 97 (1997) 1515–1566.
- [2] T. Moriuchi, M. Nishiyama, T. Hirao, A zinc(II) complex composed of a tridentate ligand bearing podand pyrenyl moieties, *Eur. J. Inorg. Chem.* (2002) 447–451.
- [3] S.-Y. Moon, N.J. Youn, S.M. Park, S.-K. Chang, Diametrically disubstituted cyclam derivative having Hg<sup>2+</sup>-selective fluoroionophoric behaviors, *J. Org. Chem.* 70 (2005) 2394–2397.
- [4] J. Kawakami, T. Niiyama, S. Ito, Intramolecular excimer formation and complexing behavior of tridentate pyridine podand having two naphthalene rings as a fluorescent chemosensor for zinc ion, *Anal. Sci.* 18 (2002) 735–736.
- [5] H. Yuasa, N. Miyagawa, M. Nakatani, M. Izumi, H. Hashimoto, A tong-like fluorescence sensor for metal ions: perfect conformational switch of hinge sugar by pyrene stacking, *Org. Biomol. Chem.* 2 (2004) 3548–3556.
- [6] D.S. Karpovich, G.J. Blanchard, Relating the polarity dependent fluorescence response of pyrene to vibronic coupling—achieving a fundamental understanding of the py polarity scale, *J. Phys. Chem.* 99 (1995) 3951–3958.
- [7] J.B. Birks, *Photophysics of Aromatic Molecules*, Wiley, London, 1970.
- [8] H.K. Cho, D.H. Lee, J.-I. Hong, A fluorescent pyrophosphate sensor via excimer formation in water, *Chem. Commun.* (2005) 1690–1692.
- [9] N.J. Van der Veen, S. Flink, M.A. Deij, R.J.M. Egberink, F.C.J.M. Van Veggel, D.N. Reinhoudt, Monolayer of a Na<sup>+</sup>-selective fluorinophore on glass: connecting the fields of monolayers and optical detection of metal ions, *J. Am. Chem. Soc.* 122 (2000) 6112–6113.
- [10] J.Y. Lee, S.K. Kim, J.H. Jung, H.S. Kim, Bifunctional fluorescent calix[4]arene chemosensor for both a cation and an anion, *J. Org. Chem.* 70 (2005) 1463–1466.
- [11] A. Corma, M.S. Galletero, H. García, E. Palomares, F. Rey, Pyrene covalently anchored on a large external surface zeolite as a selective heterogeneous sensor for iodide, *Chem. Commun.* (2002) 1100–1101.
- [12] R. Nomura, K. Yamada, T. Masuda, A chromophore-labeled poly(*N*-propargylamide): a new strategy for a stimuli-responsive conjugated polymer, *Chem. Commun.* (2002) 478–479.
- [13] J. Seixas de Melo, T. Costa, M.D.G. Miguel, B. Lindman, K. Schillén, Time-resolved and steady-state fluorescence studies of hydrophobically modified water-soluble polymers, *J. Phys. Chem. B* 107 (2003) 12605–12621.
- [14] M.D. Miguel, Association of Surfactants and Polymers Studied by Luminescence Techniques, *Adv. Colloids Interface Sci.* 89 (2001) 1–23.
- [15] M.D. Miguel, H.D. Burrows, S.J. Formosinho, B. Lindman, Fluorescence studies of polymer–surfactant association, *J. Mol. Struct.* 563 (2001) 89–98.
- [16] D.Y. Sasaki, B.E. Padilla, Dithioamide metal ion receptors on fluorescent lipid bilayers for the selective optical detection of mercuric ion, *Chem. Commun.* (1998) 1581–1582.
- [17] I. Aoki, T. Harada, T. Sakaki, Y. Kawahara, S. Shinkai, Fluorescence reading-out of the molecular-recognition process, *J. Chem. Soc. Chem. Commun.* (1992) 1341–1345.
- [18] Y. Suzuki, T. Morozumi, H. Nakamura, M. Shimomura, T. Hayashita, R.A. Bartsh, New fluorimetric alkali and alkaline earth metal cation sensors based on noncyclic crown ethers by means of intramolecular excimer formation of pyrene, *J. Phys. Chem. B* 102 (1998) 7910–7917.
- [19] Y. Nakahara, Y. Matsumi, W.B. Zhang, T. Kida, Y. Nakatsuji, I. Ikeda, Fluorometric sensing of alkaline earth metal cations by new lariat ethers having plural pyrenylmethyl groups on the electron-donating sidearms, *Org. Lett.* 4 (2002) 2641–2644.
- [20] S. Nishizawa, Y. Kato, N. Teramae, Fluorescence sensing of anions via intramolecular excimer formation in a pyrophosphate-induced self-assembly of a pyrene-functionalized guanidinium receptor, *J. Am. Chem. Soc.* 121 (1999) 9463–9464.
- [21] I. Aoki, H. Kawabata, K. Nakashima, S. Shinkai, Fluorescent calix[4]arene which responds to solvent polarity and metal ions, *J. Chem. Soc. Chem. Commun.* (1991) 1771–1772.
- [22] A. Okamoto, T. Ichiba, I. Saito, Pyrene-labeled oligodeoxynucleotide probe for detecting base insertion by excimer fluorescence emission, *J. Am. Chem. Soc.* 126 (2004) 8364–8365.
- [23] Y. Nakahara, T. Kida, Y. Nakatsuji, M. Akashi, Synthesis of double-armed lariat ethers with pyrene moieties at each end of two sidearms and their fluorescence properties in the presence of alkali metal and alkaline earth metal cations, *J. Org. Chem.* 69 (2004) 4403–4411.
- [24] F. Fages, S. Leroy, T. Soujanya, J.E.S. Sohna, Photoactive pyrene-containing receptors for transition-metal ions, *Pure Appl. Chem.* 73 (3) (2001) 411–414.
- [25] J.Y. Lee, S.K. Kim, J.H. Jung, J.S. Kim, Bifunctional fluorescent calix[4]arene chemosensor for both a cation and an anion, *J. Org. Chem.* 70 (2005) 1463–1466.
- [26] J.C. MacDonald, G.M. Whitesides, Solid-state of hydrogen-bonded tapes based on cyclic secondary diamines, *Chem. Rev.* 94 (1994) 2383–2420.
- [27] P. Jonkheijm, F.F.M. Hoebe, R. Kleppinger, J. Van Herikhuizen, A.P.H.J. Schenning, E.W. Meijer, Transfer of  $\pi$ -conjugated columnar stacks from solution to surfaces, *J. Am. Chem. Soc.* 125 (2003) 15941–15949.
- [28] X. Huang, C. Li, S. Jiang, X. Wang, B. Zhang, M. Liu, Supramolecular chirality of the hydrogen-bonded complex Langmuir–Blodgett film of achiral barbituric acid and melamine, *J. Colloids Interface Sci.* 285 (2005) 680–685.
- [29] P.V. Bernhardt, E.J. Hayes, Redox active macrocyclic receptors for neutral guests, *Inorg. Chem.* 42 (2003) 1373–1377.
- [30] N.A. Bailey, D.E. Fenton, S.J. Kitchen, T.H. Lilley, M.G. Williams, P.A. Tasker, A.J. Leong, L.F. Lindoy, Metal-ion selectivity by macrocyclic ligands. Part 1: the interaction of Ni<sup>II</sup> and Cu<sup>II</sup> with pyridinyl-derived N<sub>3</sub>O<sub>2</sub> macrocycles. The X-ray structures of a free macrocycle, its endo-macrocyclic complexes of Ni<sup>II</sup> and Cu<sup>II</sup> and exomacrocyclic nickel(II) complex, *J. Chem. Soc. Dalton Trans.* (1991) 627–637.
- [31] J. Seixas de Melo, P.F. Fernandes, Spectroscopy and photophysics of 4- and 7-hydroxycoumarins and their thione analogs, *J. Mol. Struct.* 565 (2001) 69–78.
- [32] G. Striker, V. Subramaniam, C.A.M. Seidel, A. Volkmer, Photochemistry and fluorescence lifetimes of green fluorescent protein, *J. Phys. Chem. B* 103 (1999) 8612–8617.
- [33] J.L. Capelo, A.V. Figueiras, I. Lavilla, C. Bendicho, Solid–liquid extraction of copper from slurried samples using high intensity probe sonica-

tion for electrothermal atomic absorption spectrometry, *Talanta* 50 (1999) 905–911.

- [34] (a) C. Lodeiro, R. Bastida, A. de Blás, D.E. Fenton, A. Macías, A. Rodríguez, T. RodríguezBlas, Complexes of lead(II) and lanthanide(III) ions with two novel 26-membered-imine and -amine macrocycles derived from 2,6-bis(2-formylphenoxy)methylpyridine, *Inorg. Chim. Acta* 267 (1998) 55–62;
- (b) E. Bértolo, R. Bastida, A. De Blás, D.E. Fenton, C. Lodeiro, A. Macías, A. Rodríguez, T. Rodríguez-Blas, Lanthanide(III) nitrate complexes of two 17 membered N<sub>3</sub>O<sub>2</sub>-donor macrocycles, *J. Incl. Phen. Mac. Chem.* 35 (1999) 191–198;
- (c) C. Lodeiro, R. Bastida, E. Bértolo, A. Macías, A. Rodriguez, Coordination chemistry of copper(II) with oxaza macrocyclic ligands: crystal structure of a dinuclear tetramer copper(II) complex, *Polyhedron* 22 (2003) 1701–1710;
- (d) C. Lodeiro, R. Bastida, E. Bértolo, A. Macías, A. Rodríguez, Metal complexes with four macrocyclic ligands derived from 2,6-bis(2-formylphenoxy)methylpyridine and 1,7-bis(2'-formylphenyl)1,4,7-trioxaheptane, *Inorg. Chim. Acta* 343 (2003) 133–140;
- (e) C. Lodeiro, R. Bastida, E. Bértolo, A. Rodriguez, A new family of N<sub>x</sub>O<sub>y</sub> pyridine-containing macrocycles: synthesis and characterization of their Y(III), Ln(III), Zn(II), and Cd(II) coordination compounds, *Can. J. Chem. Rev. Can. Chim.* 82 (2004) 437–447.
- [35] S.C. Cummings, D.H. Busch, Geometric and optical isomers of cobalt(III) complexes containing 2 moles of cyclic tridentate ligand derived from ortho aminobenzaldehyde, *J. Am. Chem. Soc.* 92 (1970) 1924–1970.
- [36] S.M. Peng, G.C. Gordon, V.L. Goedken, Template condensations: metal-ion-directed synthesis of macrocyclic complexes from 2,3-butanodione dihydrazone and aldehydes or ketones, *Inorg. Chem.* 17 (1978) 119–126.
- [37] N.S. Gill, R.H. Nuttall, D.E. Scaife, The infra-red spectra of pyridine complexes and pyridinium salts, *Inorg. Nucl. Chem.* 18 (1961) 79–87.
- [38] S. Aime, M. Botta, U. Casellato, S. Tamburini, P.A. Vigato, NMR evidence for interconversion between 2 enantiomeric forms of macrocyclic Schiff-base lanthanide(III) complexes through reversible ring contraction and expansion, *Inorg. Chem.* 34 (1995) 5825–5831.
- [39] K. Nakamoto, *Infrared and Raman Spectra of Inorganic and Coordination Compound*, third ed., Wiley-Interscience, New York, 1978.
- [40] F. Pina, J.C. Lima, C. Lodeiro, J. Seixas de Melo, P. Diaz, M.T. Albelda, E. Garcia-Espana, Long range electron transfer quenching in polyamine chains bearing a terminal naphthalene unit, *J. Phys. Chem. A* 106 (2002) 8207–8212.
- [41] A.P. de Silva, H.Q.N. Gunaratne, C.P. McCoy, Direct visual indication of pH windows: 'off-on-off' fluorescent PET (photoinduced electron transfer) sensors switches, *Chem. Commun.* (1996) 2399–2400 (references therein).
- [42] J. Seixas de Melo, J. Pina, F. Pina, C. Lodeiro, A.J. Parola, J.C. Lima, M.T. Albelda, M.P. Clares, E. García-España, Energetics and dynamics of naphthalene polyaminic derivatives: influence of structural design in the balance static vs dynamic excimer formation, *J. Phys. Chem. A* 107 (2003) 11307–11318.
- [43] S. Draxler, M.E. Lippitsch, pH sensors using fluorescence decay time, *Sens. Actuators B* 29 (1995) 199–203.
- [44] Since experiment was carried out at pH 4 in 99.5% water–0.5% acetonitrile what we have considered is the interaction between the protonated barbituric acids and the diprotonated form of the ligand that exist at this pH. The pK<sub>a</sub> values of cyanuric acid (6.88) [45] and parabanic acid (5.9) [46] show that proton transfer to the ligand that not occurred at pH 4.
- [45] (a) E.M. Smolin, L. Rapoport, *The Chemistry of Heterocyclic Compounds*, vol. 13, Interscience, New York, 1959, pp. 33–36;
- (b) L.J. Prins, D.N. Reinhoudt, P. Timmerman, Noncovalent synthesis using hydrogen bonding, *Angew. Chem. Int. Ed.* 40 (2001) 2382–2426.
- [46] A.E. Martell, R.M. Smith, NIST Critically selected stability constants of metal complexes, NIST Standard Reference Database 46, Version 6.0, Texas A&M University, USA, 2001.

## Biographies



**C. Lodeiro** received his PhD in chemistry in 1999 from Santiago de Compostela University, Spain. In 1999, he moved to the New University of Lisbon as a post-doc working in a European project concerning molecular devices and machines. In 2004, he became a fellow researcher of REQUIMTE, CQFB and invited assistant professor (lecturer) at the Chemistry Department, New University of Lisbon, Portugal. He is member of the Portuguese and the Royal Spanish Chemistry Societies. His research interests comprise organic and inorganic synthesis, macrocyclic systems, supramolecular photochemistry, chemical sensors and green synthetic methods.



**J.C. Lima** received his PhD in chemistry from the Technical University of Lisbon, IST in 1996. He was appointed assistant professor in the New University of Lisbon in 2000, teaching inorganic chemistry, photochemistry, chemistry structure and bonding, and physical chemistry at the Department of Chemistry of the New University of Lisbon. He is a member of the Portuguese Chemical Society. His research interests concern fluorescence of proteins, flavylum salts with different substitution patterns and fluorescent chemosensors. He is a member (researcher) of

REQUIMTE (a national associated laboratory denoted to research in green chemistry).



**A.J. Parola** received his PhD in chemistry from the New University of Lisbon in 1998. He was then appointed assistant professor, teaching inorganic chemistry and physical chemistry at the Department of Chemistry of the New University of Lisbon. He is a member of the Portuguese Chemical Society. His research interests concern supramolecular photochemistry, from the design and synthesis of supramolecular species to the study of their photo-physical properties, in particular, photoactive molecular cages (hemiacararands), flavylum salts with different substitution patterns and fluorescent chemosensors. He is a member

(researcher) of REQUIMTE.



**J. Sérgio Seixas de Melo** received his PhD in chemistry from the Technical University of Lisbon (IST), Portugal, in 1996. In 1993, he was appointed assistant lecturer in the University of Coimbra, Portugal, after which (1998) he became assistant professor (lecturer) in the same university. He still holds this position, and lectures there in the courses of general chemistry and photochemistry. He is an active member of the Portuguese Chemistry Society. His main research interests deal with the study of the excited states of polymers (organic conjugated and hydrophobically modified), photophysics of "molecules of art", molecular devices and kinetic processes in the excited state.



**J.L. Capelo** obtained his PhD in chemistry in 2002 from Vigo University, Spain, where he has been an associated professor (lecturer) from 1999 to 2002. In 2002, he moved to IST, Technical University of Lisbon, as a post-doc. In 2005, he became a fellow researcher of REQUIMTE, CQFB, at the Chemistry Department, New University of Lisbon. He is member of the Portuguese Chemistry Society. His research interests comprise analytical chemistry, new green analytical methods, AOPs, ultrasound and microwave techniques.



**A. Tamayo** obtained his MSc degree in 2001 in chemistry in the Autonomia University of Barcelona, Spain, and is currently, a PhD student in Barcelona (UAB). He moved to the Chemistry Department of the Nova University of Lisbon during a short period of 4 months between 2004 and 2005. He is interested in supramolecular chemistry, organic and inorganic synthesis.



**B. Covelo** obtained her PhD in chemistry in 2004 from Vigo University, Spain. Actually, she is a post-doc in the Photochemistry and Supramolecular Chemistry Group of REQUIMTE, CQFB, Chemistry Department, Nova University of Lisbon, Portugal. Her research interests include inorganic chemistry, photochemistry, supramolecular chemistry and X-ray diffraction.



**B. Pedras** obtained his MSc degree from the Lisbon Faculty of Science (FCL), Portugal in 2004 in chemistry, and is currently a research student in the Nova University of Lisbon (FCT-UNL). He is interested in supramolecular chemistry, organic and inorganic synthesis.