Tetrahedron Letters 50 (2009) 6383–6388

Contents lists available at ScienceDirect

Tetrahedron Letters

journal homepage: www.elsevier.com/locate/tetlet

Dual Bodipy fluorophores linked by polyethyleneglycol spacers

Soumyaditya Mula [a,b], Gilles Ulrich [a], Raymond Ziessel [a,*]

[a] *Laboratoire de Chimie Organique et Spectroscopies Avancées (LCOSA), Ecole Chimie Polymères, Matériaux (ECPM), 25 rue Becquerel, 67087 Strasbourg Cedex, France*
[b] *Bio-Organic Division, Bhabha Atomic Research Centre, Mumbai 400 085, India*

ARTICLE INFO

Article history:
Received 29 June 2009
Revised 31 July 2009
Accepted 26 August 2009
Available online 1 September 2009

Keywords:
Bodipys'
Dual dyes
Amino groups
Polyethyleneglycol
Fluorescence
Sensors

ABSTRACT

Several Bodipy dyes were synthesized with various substituents designed to potentially interact with adventurous cations including protons. Among the different synthetic strategies, protection of the amino group by a BOC appears efficient for the substitution of the fluoro groups on the boron by Grignard reagents. Single alkylation on the amino-Bodipy compounds by a polyethyleneglycol chain bearing an alkyne end group is feasible using a biphasic strategy. Linkage of a blue dye on the alkyne fragment is promoted by palladium(0) and it provides novel dual donor–acceptor dyes. Fluorescence of the mono-alkylated amino dyes is heavily quenched but restored by protonation or interaction with Fe(II) salts. Very efficient energy transfer from the donor (energy input at 20,000 $\mathrm{cm^{-1}}$) to the acceptor (energy output at 15,100 $\mathrm{cm^{-1}}$) is quantitative and not distance dependent.

Linked fluorophores are interesting artificial models for mimicking energy transfer processes, antenna effects, photon concentration and charge separation occurring in photosynthetic organisms and plants.[1] There is also current interest in such molecules for use as ratiometric dyes for sensing analytes,[2] and in cascade energy transfer events related to photon concentration and chemical transformations.[3,4] The fundamental event involved in such systems is Förster resonance energy transfer (FRET), which relies on judicious selection of the absorption and emission properties of the dyes and the way they are oriented and linked to the central structural platform.[5] Recently, several systems have been developed in which arrays of sophisticated multi-chromophoric dyes ensure very fast multi-energy transfer processes, so that all absorbed photons are concentrated spatially to one single emitting dye.[6–10]

These systems are fascinating due to the synthetic challenges associated with the need for selective cross-couplings, judicious choice of the linking functions and careful purification of intermediates. We have found that the use of unsaturated linkages facilitates the synthesis of the building blocks as well as the assemblage of the final molecules with shapes and functions related to molecular scale wires, dendrimers, tandem scaffoldings and cassette edifices.[11]

The main results obtained so far and scrutinized in several reviews concern: (i) the ratiometric sensing of cations and protons in solution and cellular environments, (ii) the amplification of Stokes shifts and (iii) the engendering of very efficient excitonic energy transfer approaching the efficiency of natural systems.[12,13]

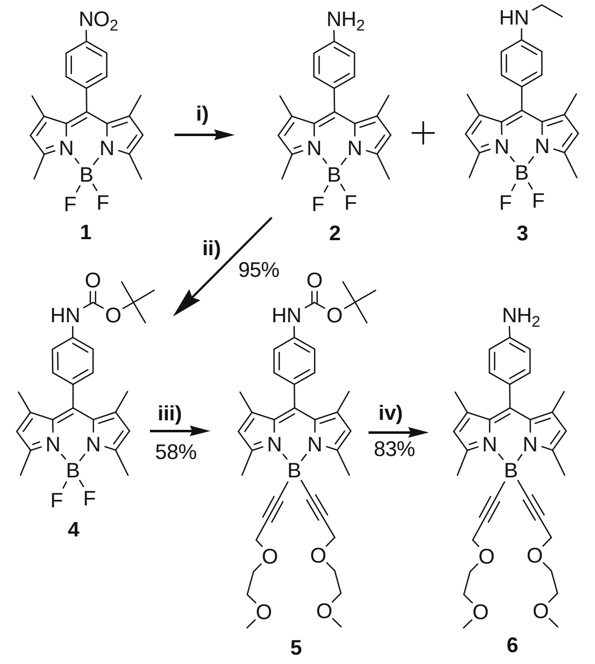
The concept exploited herein is to link two dyes, both derivatives of difluoro-boradiaza-s-indacene, commonly termed Bodipy (for boron dipyrromethene),[12,13] having two different colours and linked by a flexible bridge (polyethyleneglycol) in order to favour FRET between the dyes, a process enhanced by folding the dyes in the presence of cations. The system has been designed in such a way that the dye emitting at higher energy (green emitter) is fluorescent solely in the presence of proton or cations, due to the fact that the fluorescence of amino-Bodipy is effectively quenched by photoinduced electron transfer or more likely by a localized charge transfer state. Our first choice was targeted on such derivatives carrying suitable motifs for absorption and emission tunings, with solubilizing groups attached on the boron to avoid aggregation problems (Scheme 1).[14]

Compound **2** can be easily obtained from **1** by Pd-catalyzed hydrogenation, as reported previously,[15] but it requires 24 h to complete the reaction (Scheme 1) which is disadvantageous when the dye possesses a reducible group like a double bond, as was found to be the case when starting with the distyryl derivative **7**, where we ended up with an inseparable mixture of compounds. After some experimentation, we found that Pd-catalyzed hydrogenation in an ethanol/acetonitrile mixture at 70 °C was a good alternative as it took only 15 min to complete the reduction (Scheme 1). In addition to its rapidity, in this method it is possible to synthesize the *N*-ethylamino dye[16] which is a very useful precursor for the preparation of unsymmetrical tertiary amino-Bodipy dyes. With a prolonged reaction time (24 h) under the same conditions,

---

[*] Corresponding author. Tel./fax: +33 3 68 85 2761.
E-mail address: ziessel@unistra.fr (R. Ziessel).

0040-4039/$ - see front matter © 2009 Elsevier Ltd. All rights reserved.
doi:10.1016/j.tetlet.2009.08.091



Scheme 1. Reagents and conditions: (i) see Table 1; (ii) di-*tert*-butyldicarbonate, dry THF, 60 °C, 2 days; (iii) $CH_3OCH_2CH_2OCH_2C{\equiv}CMgBr$, dry THF, 60 °C, 12 h; (iv) 6% TFA, dry CH_2Cl_2, 2 h.

Table 1
Reaction conditions and synthetic outline

Reagents key (i)	Time	Ratio 2 versus Ratio 3
H_2, 10% Pd/C (10%), three drops of water, CH_2Cl_2/EtOH (1:1), rt	24 h	100:0
H_2, 10% Pd/C (10%), three drops of water, EtOH/CH_3CN (1:1), 70 °C	15 min	95:5
H_2, 10% Pd/C (10%), three drops of water, EtOH/ CH_3CN (1:1), 70 °C	24 h	15:85

compound **3** was obtained in very good yield at the expense of **2** (Table 1).

One of the most critical aspects of *Bodipy* dye chemistry is control of their solubility, which is very important for their large scale preparation. The boron centre provides a convenient site to attach a functional group without affecting the optical properties of the chromophore.[17] Furthermore, the use of 2,5-dioxaoctyne is a convenient fragment which imports solubility, polarity and processability to the dye. It has been found that 8-(*N*-Boc)-anilino *Bodipy* **4**, synthesized from **2** via the reaction with BOC anhydride (Scheme 1), is stable enough to react with an alkynyl Grignard[18] to substitute the fluoro groups on boron centre, and **5** was isolated in reasonably good yield (58%). Acid cleavage of the *N*-Boc with 6% TFA then furnished the highly soluble dye **6** functionalized at the boron centre. By syntheses analogous to those of the yellow amino derivatives **2** and **6**, we prepared the blue amino analogues **8** and **12**. The distyryl compound **7** was synthesized from **1** by a Knoevenagel condensation in good yield.[19,20] The reduction of **7** using the previous conditions did produce the desired amino derivative **8**, with no ethylamino derivative being isolated, but a non-conjugated yellow dye **9** was also isolated in 5% yield (Table 2) as a result of over-reduction of the double bonds.

Table 2
Reaction conditions and synthetic outline

Reagent key (ii)	Time (h)	Ratio 8 versus Ratio 9
H_2, 10% Pd/C (10%), three drops of water, EtOH/CH_3CN (1:1), 70 °C	4	95:5
H_2, 20% Pd/C (10%), three drops of water, EtOH/CH_3CN (1:1), 70 °C	2	0:100

After BOC-protection of the amino group of **8**, reaction at the boron centre with the alkynyl Grignard furnished the soluble compound **11**. The final amino-*Bodipy* **12** with a different emission wavelength and improved solubility was obtained after *N*-Boc cleavage by TFA (see Scheme 2).

The nucleophilic character of the amino function is unfortunately less reactive than related systems, such as *p*-nitrophenylsulfonamide-protected amino groups.[21] In our case, several efforts to substitute the amino function in **6** and **12** were unsuccessful. So we decided to keep the fluoride on the Boron centre and to solve the solubility problem in a different fashion. Thus, we tried substitution reactions at the amino centres of **2** and **8** using different, protected polyethyleneglycol-based substrates. Biphasic reaction in water using $NaHCO_3$ as the base was found to be the best strategy (Scheme 3).[22] Though the yield was moderate, the reaction furnished exclusively the mono-N-substituted product **13** with a linker suitable for subsequent substitution. Unfortunately this biphasic reaction of product **13** with the blue amino-derivatives **8** or **12** was ineffective probably because of the weaker nucleophilicity of the amino function in this delocalized dye.

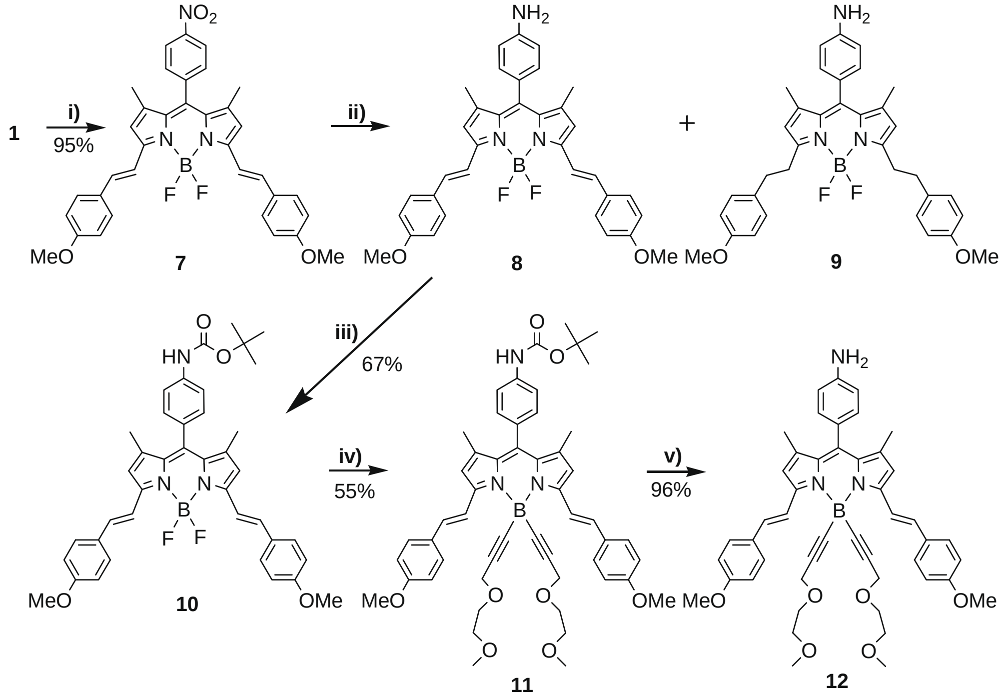
In order to reach the target we chose to prepare an unsymmetrical linker bearing on one side a tosylate (activable with derivative **2**) and on the other an ethynyl substituent able to be cross-linked conveniently using routine palladium catalysis with the iodo-substituted blue dye **15**. The hybrid linkers were prepared from the ditosylate of polyethylene glycol with propargylic alcohol under basic conditions (Scheme 4).

Substitution of the TsO fragment by derivative **2** was then straightforward, affording the green emitters **14a–c** in acceptable yields (Scheme 5). To explore the synthetic utility of compounds **14**, we cross-coupled 4,4-difluoro-3,7-di-*p*-methoxystyryl-1,7-dimethyl-8-*p*-iodophenyl-4-bora-3a,4a-diaza-s-indacene (**15**) via a Sonogashira reaction promoted by Pd(0) to furnish **16a–c** in satisfactory yields (Scheme 5). Finally, we also succeeded in coupling dye **14a** with 1-bromopyrene **17** to get the linked derivative **18** absorbing at about 380 nm (Scheme 6).

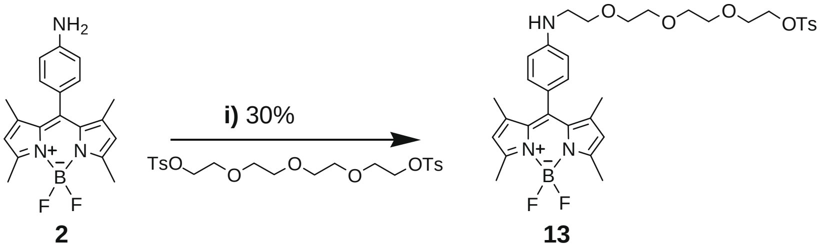
Preliminary electrochemical measurements for **3**, **14a**, **15** and **16a** are given in Table 3. For all these dyes, a single reversible cathodic wave was observed due to the (*Bodipy/Bodipy*$^{\cdot -}$) couple. In the case of **15**, the reduction was facilitated by ~300 mV as compared to dyes **3** and **14a**. Anodically, an irreversible wave was observed along with a reversible wave for (Bodipy/Bodipy$^{\cdot +}$). The irreversible wave for **14a** was due to the oxidation of the secondary amine centre and was also seen for **3**, but not for the 8-amino derivatives. Alkyl substitution made the amine centre easily oxidizable. As expected, the anodic behaviour of **15** was more complicated than that of the other dyes. Two extra waves were observed at higher potential and were tentatively assigned to oxidation of the styryl entity and a second oxidation of the *Bodipy* core.

Interestingly, all the characteristic anodic and cathodic waves for both the precursor dyes **14a** and **15** were found in the final dye **16a**. In particular, two reversible cathodic waves separated by 290 mV were found for the (*Bodipy/Bodipy*$^{\cdot -}$) couples for both the dye cores. As expected, the anodic part was more complicated due to the overlap of the oxidation waves of the amine and styryl units. These data are in keeping with a dual dye where both chromophores/luminophores interact weakly.

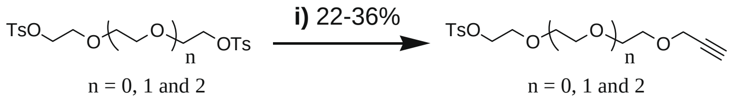
The amino derivatives **2** and **8** gave similar fluorescence quantum yields (QY = 32–38%), whereas substitution by a BOC-protecting group in **4** and **10** increased the QYs to 84% and 49%,



Scheme 2. Reagents and conditions: (i) *p*-methoxybenzaldehyde, piperidine, toluene, reflux; (ii) H_2, Pd/C (10%), three drops of water, EtOH/CH_3CN (1:1), 70 °C; (iii) BOC anhydride, dry THF, 60 °C, 2 days; (iv) $CH_3OCH_2CH_2OCH_2C{\equiv}CMgBr$, dry THF, 60 °C, 12 h; (v) 6% TFA, dry CH_2Cl_2, 2 h.



Scheme 3. Reagents and conditions: (i) $NaHCO_3$, dodecyl sulfate, $C_2H_4Cl_2$, H_2O, 80 °C, 24 h.

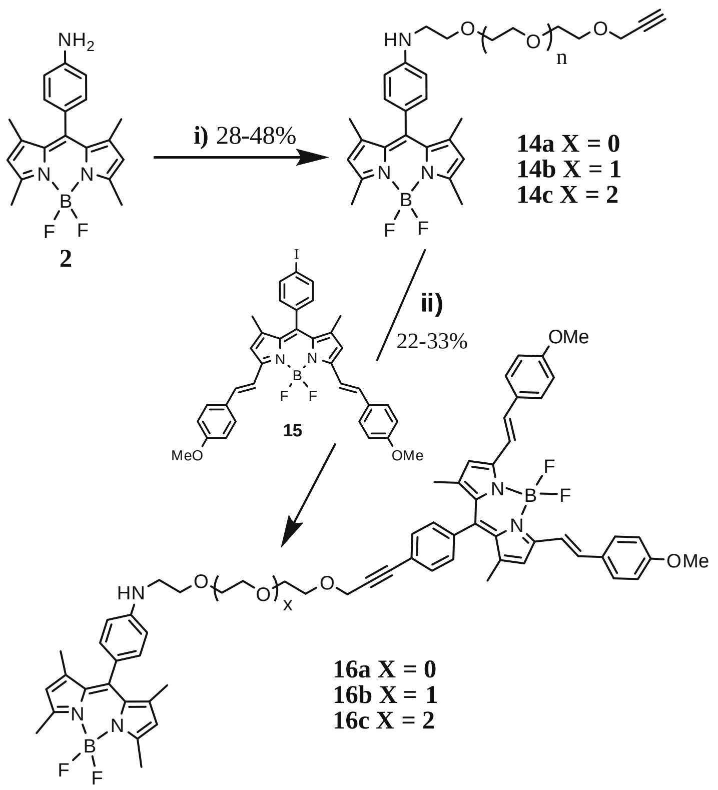


Scheme 4. Reagents and conditions: (i) propargylic alcohol (1.2 equiv, *t*-BuOK, THF).

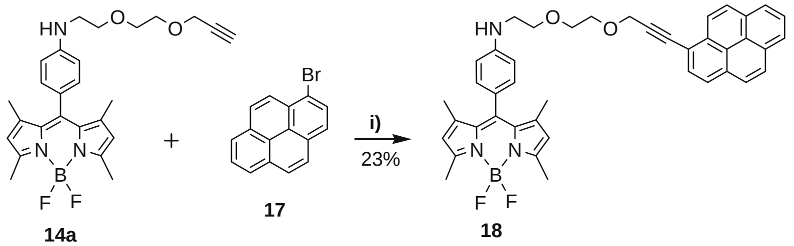
respectively. Alkylation of the amino function by alkyl fragments as in **3** (with ethyl) and **14a–c** (with polyethyleneglycol) resulted in a dramatic decrease of the QY to 2–3%, while the excited state lifetimes remained similar (Table 4). It is likely that the electron-donating fragment (NHR) and the electron-accepting *Bodipy* favour the emergence of a localized charge transfer state responsible for the marked quenching of the fluorescence. This non-emissive state might however favour fast energy transfer from the green emitter to the blue acceptor. Interestingly, in the dual dyes **16a–c** where the absorption spectra are simple sums of the separate dye spectra (Fig. 1), the electronic energy transfer is almost quantitative, despite the weak fluorescence of the model derivatives **14a–c**, and is not dependent on the distance between the two dyes.

For the pyrene-linked dye **18**, excitation in the pyrene residue at 362 nm resulted in very weak pyrene fluorescence at 393 nm (<0.2%) but higher fluorescence of S_1–S_0 transition of the *Bodipy* unit at 516 nm (Fig. 2). A large weak broad emission band centred at 650 nm which could be attributed to a low lying charge transfer emissive state is also observed.[24] Protonation of the amino moiety in **18** using gaseous HCl resulted in a QY increase from 2% to 40%, whereas spicing the solution with Fe^{2+} salts resulted in an increase to 33% (Fig. 3). Iron(II) has a strong affinity for amino groups and is relatively oxophile and might consequently strongly interact with the amino-polyethyleneglycol spacer.

From a mechanistic viewpoint, this is an interesting situation where the fluorescence of the donor can be switched on by protons or Fe^{2+} (selectively versus other metallic salts) and a classical FRET is then effective, favoured by the spectral overlap between the emission of the green emitter and the absorption of the blue dye. In the absence of cations, it is possible that a charge transfer state effectively favours EET without the need to channel through the emission of the donor.



Scheme 5. Reagents and conditions: (i) $NaHCO_3$, dodecyl sulfate, corresponding tosylate, $C_2H_4Cl_2$, H_2O, 80 °C, 24 h; (ii) $[Pd(PPh_3)_4]$, benzene, Et_3N, 50 °C.



Scheme 6. Reagents and conditions: (i) $[Pd(PPh_3)_4]$, benzene, $(iPr)_2NH$, 70 °C.

In short, new borondipyrromethene dyes carrying amino-linked derivatives have been developed and dual-Bodipy species have been engineered displaying both an orange-red colour (with emission at 540 nm) and a blue colour (emission at 650 nm). The method established for the synthesis of new dyads in which orange and blue dyes are linked via various polyethyleneglycol chains has provided families of compounds where, in one case, the blue dye has been replaced by a pyrene residue. Fluorescence measurements revealed quenching processes inherent to the presence of the amino fragments and probably due to the presence of a charge transfer state which might be responsible for the efficient energy transfer between the emitting states of the orange and blue species. Interactions with protons and cations have also been evaluated and more detailed mechanistic studies are currently in progress.

Table 3
Properties of selected dyes[a]

Compd	E^0_{ox} V (ΔE, mV)	E^0_{red}, V (ΔE, mV)
3	+1.04 (irrev); +1.21 (90)	−1.30 (70)
14a	+0.98 (irrev); +1.12 (90)	−1.27 (70)
15	+0.74 (60); +1.08 (70); +1.58 (irrev)	−0.97 (60); −1.90 (irrev)
16a	+0.72 (60); +0.98 (irrev); +1.11 (irrev); +1.22 (irrev); +1.57 (irrev)	−1.01 (70); −1.29 (74); −1.88 (irrev)

[a] Potential determined by cyclic voltammetry in deoxygenated CH_2Cl_2 solutions, containing 0.1 M $TBAPF_6$, at a solute concentration range 1–5 × 10^{-3} M, at 23 °C. Potentials were standardized with ferrocene (Fc) as internal reference and converted to SSCE assuming that $E_{1/2}$ (F_c/F_c^+) = 0.38 V (ΔE_p = 70 mV) SSCE. The error in half-wave potentials was ±10 mV, where the redox process was irreversible and the peak potential ($E_{ap/cp}$) was quoted. All waves were monoelectronic unless otherwise specified.

Table 4
Optical properties of selected compounds at rt[a]

Compd	λ_{abs} (nm)	ε_{max} (M^{-1} cm^{-1})	λ_{em} (nm)	Φ_F (%)[b]	τ_{em} (ns) ±0.01	k_r (10^8 s^{-1})[c]	k_{nr} (10^6 s^{-1})[c]
2	500	80,000	510	38	3.14	1.21	197.4
3	499	86,000	513	2	3.00	0.07	326.7
4	501	86,000	510	84	3.32	2.53	48.2
5	499	81,000	507	85	3.27	2.60	15.3
6	500	79,000	509	34	2.98	0.80	221.5
8	641	102,000	656	32	5.75	0.56	118.3
	368	69,000					
9	504	99,000	514	38	3.04	1.25	203.9
10	642	99,000	658	49	5.67	0.86	89.9
	369	62,000					
11	642	90,000	656	54	5.49	0.98	83.8
	369	52,000					
12	640	68,000	659	40	5.79	0.69	103.6
	367	41,000					
14a	500	103,000	515	3	3.12	0.01	310.9
14b	500	98,000	515	3	2.95	0.10	328.8
14c	500	74,000	514	3	3.00	0.10	323.3
16a	646	120,000	668	39	7.01	0.56	87.0
	500	86,000	514	1			
	372	80,000					
16b	646	112,000	668	38	5.97	0.64	103.8
	501	77,000	514	0.7			
	372	74,000					
16c	646	107,000	662	42	5.81	0.72	99.8
	500	73,000	512	0.8			
	371	71,000					
18	500	77,000	516	2	4.46	0.04	220.6
	362	42,000	393	0.2			

[a] Determined in anhydrous CH_2Cl_2 solution.

[b] Determined using rhodamine 6G, as reference $\Phi = 0.78$ in water, $\lambda_{exc} = 488$ nm [Ref. 23], all Φ_F are corrected for changes in refractive index.

[c] Calculated using the following equations: $k_r = \Phi_F/\tau_F$ and $k_{nr} = (1 - \Phi_F)/\tau_F$.

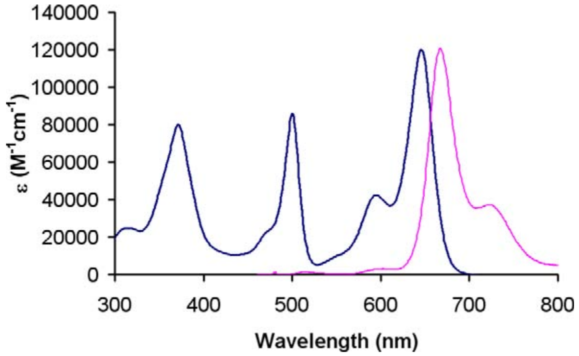


Figure 1. Absorption (blue) and fluorescence (pink) spectra of **16a** in dichloromethane at rt.

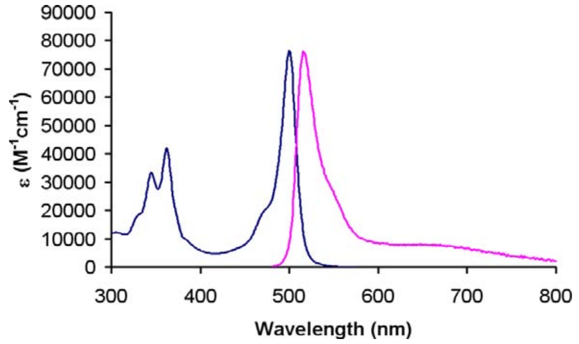


Figure 2. Absorption (blue) and fluorescence (pink) spectra of **18** in dichloromethane at rt.

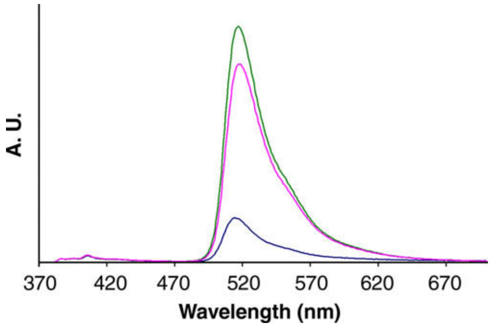


Figure 3. Fluorescence spectra of **18** (blue), **18** + H^+ (green) and **18** + Fe^{2+} (pink), $\lambda_{exc} = 480$ nm in dichloromethane at rt. Acid was introduced as vapours and the iron salt in a methanol solution.

Acknowledgements

This work was partially supported by the CNRS providing research facilities. Soumyaditya Mula is very grateful to the French Embassy in India for the award of a Sandwich-Ph.D. Scholarship and also to the Bhabha Atomic Research Centre for permission to conduct research in France. We also warmly thank Professor Jack Harrowfield (ISIS in Strasbourg) for commenting on the manuscript before publication.

References and notes

1. Blankenship, R. E. In *Molecular Mechanism of Photosynthesis*; Blackwell Science: Oxford, UK, 2002.
2. Coskun, A.; Akkaya, E. U. *J. Am. Chem. Soc.* **2006**, *128*, 14474–14475.
3. Harriman, A.; Mallon, L.; Ziessel, R. *Chem. Eur. J.* **2008**, *14*, 11461–11473.
4. Harriman, A.; Mallon, L.; Goeb, S.; Ulrich, G.; Ziessel, R. *Chem. Eur. J.* **2009**, *15*, 4553–4564.

5. (a) Förster, T. *Ann. Phys.* **1948**, *2*, 55–75; (b) Stryer, L.; Haughland, R. P. *Proc. Natl. Acad. Sci.* **1967**, *58*, 719–726.
6. Holten, D.; Bocian, D. F.; Lindsey, J. S. *Acc. Chem. Res.* **2002**, *35*, 57–69.
7. Serin, J. M.; Brousmiche, D. W.; Fréchet, J. M. J. *Chem. Commun.* **2002**, 2605–2607.
8. (a) Weil, T.; Reuther, E.; Müllen, K. *Angew. Chem., Int. Ed.* **2002**, *41*, 1900–1904; (b) Cotlet, M.; Vosch, T.; Habichi, S.; Weil, T.; Müllen, K.; Hofkens, J.; De Schryver, F. *J. Am. Chem. Soc.* **2005**, *127*, 9760–9768.
9. Yuan, M.; Yin, X.; Zheng, H.; Ouyang, C.; Zuo, Z.; Liu, H.; Li, Y. *Chem. Asian J.* **2009**, *4*, 707–713.
10. (a) Diring, S.; Punteriero, F.; Nastasi, F.; Campagna, S.; Ziessel, R. *J. Am. Chem. Soc.* **2009**, *131*, 6108–6110; (b) Haefele, A.; Ulrich, G.; Retailleau, P.; Ziessel, R. *Tetrahedron Lett.* **2008**, *49*, 3716–3721.
11. Harriman, A.; Ziessel, R. In *Carbon-Rich Compounds*; Haley, M. M., Tykwinski, R. R., Eds.; Wiley-VCH: Weiheim, 2006; pp 26–89. and references cited therein.
12. (a) Ziessel, R. *C.R. Chimie* **2007**, *10*, 622–629; (b) Ulrich, G.; Ziessel, R.; Harriman, A. *Angew. Chem., Int. Ed.* **2008**, *47*, 1184–1201; (c) Ziessel, R.; Ulrich, G.; Harriman, A. *New J. Chem.* **2007**, *31*, 496–501.
13. Loudet, A.; Burgess, K. *Chem. Rev.* **2007**, *107*, 4891–4932.
14. Niu, S.-L.; Ulrich, G.; Ziessel, R.; Kiss, A.; Renard, P.-Y.; Romieu, A. *Org. Lett.* **2009**, *11*, 2049–2052.
15. Ziessel, R.; Bonardi, L.; Retailleau, P.; Ulrich, G. *J. Org. Chem.* **2006**, *71*, 3093–3096.
16. (a) Sajiki, H.; Ikawa, T.; Hirota, K. *Org. Lett.* **2004**, *6*, 4977–4980; (b) Nakario, R.; Kotakonda, S.; Fouchard, D. M. D.; Tillekeratne, L. M. V.; Hudson, R. A. *Org. Lett.* **2005**, *7*, 471–474.
17. (a) Goze, C.; Ulrich, G.; Ziessel, R. *J. Org. Chem.* **2007**, *72*, 313–322; (b) Ulrich, G.; Goeb, S.; De Nicola, A.; Retailleau, P.; Ziessel, R. *Synlett* **2007**, 1517–1520; (c) Bonardi, L.; Ulrich, G.; Ziessel, R. *Org. Lett.* **2008**, *10*, 2183–2186.
18. Zhang, P.; Terefenko, E. A.; Slavin, J. *Tetrahedron Lett.* **2001**, *42*, 2097–2099.
19. (a) Haughland, R. P.; Kang002C H. C. U.S. Patent 4,774,339, Sep. 27, 1988.; (b) Rurack, K.; Kollmansberger, M.; Daub, J. *New J. Chem.* **2001**, *25*, 289–292; (c) Saki, N.; Dinc, T.; Akkaya, E. U. *Tetrahedron* **2006**, *62*, 2721–2725; (d) Yu, Y.-H.; Descalzo, A. B.; Shen, Z.; Röhr, H.; Liu, Q.; Wang, Y.-W.; Spieles, M.; Li, Y.-Z.; Rurack, K.; You, X.-Z. *Chem. Asian J.* **2006**, *1–2*, 176–187.
20. Ziessel, R.; Ulrich, G.; Harriman, A.; Alamiry, M. A. H.; Stewart, B.; Retailleau, P. *Chem. Eur. J.* **2009**, *15*, 1359–1369.
21. Koutaka, H.; Kosuge, J.-I.; Fukasaku, N.; Hirano, T.; Kikuchi, K.; Urano, Y.; Kojima, H.; Nagano, T. *Chem. Pharm. Bull.* **2004**, *52*, 700–715.
22. Singh, C. B.; Kavala, V.; Samal, A. K.; Patel, B. K. *Eur. J. Org. Chem.* **2007**, 1369–1377.
23. Olmsted, J., III *J. Phys. Chem.* **1979**, *83*, 2581–2584.
24. Kollmannsberger, M.; Rurack, K.; Resch-Genger, U.; Daub, J. *J. Phys. Chem. A* **1998**, *102*, 10211–10220.