New insights into the solubilization of Bodipy dyes

Song-Lin Niu [a], Gilles Ulrich [a,*], Pascal Retailleau [b], Jack Harrowfield [c], Raymond Ziessel [a,*]

[a] *Laboratoire de Chimie Organique et Spectroscopies Avancées (LCOSA), ECPM 25 rue Becquerel 67087 Strasbourg Cedex, France*
[b] *Laboratoire de Cristallochimie, ICSN-CNRS, Bât 27-1 Avenue de la Terrasse, 91198 Gif-sur-Yvette, Cedex, France*
[c] *ISIS 8 allée Gaspard Monge BP 70028 F-67083 Strasbourg Cedex, France*

A R T I C L E   I N F O

A B S T R A C T

Several *Bodipy* dyes were synthesized with various substituents designed to improve the water solubility. Among the different synthetic strategies protection of sulfonate groups by pyrrole of indopyrrole appears efficient but the deprotection step does not offer viable routes. Conversion of the bromopyrene or iodo Bodipy compounds to sulfobetaine derivatives is feasible either by cross-coupling directly the ethynyl-sulfobetaine or by first cross-coupling 1-(*N*,*N*-dimethylamino)-prop-2-yne followed by quaternization of the dimethylamino residue with 1,3-propanesultone. Some of these dyes (Bodipy's and pyrene) are reasonably soluble in water and remain highly fluorescent in polar solvents and water.

Perhaps one of the most active areas of current research relating to fluorescent dyes is that concerning the introduction of various functionalities to improve their utility without significantly modifying the intrinsic spectroscopic properties of the dyes. Cyanines, in particular, are currently under intense investigation,[1,2] the family of difluoro-boradiaza-*s*-indacenes, which are commonly termed *Bodipy* (for boron dipyrromethene) dyes, being especially promising because of their exceptional spectroscopic properties.[3,4] These include high extinction coefficients, sharp emission bands, high quantum yields and behaviour as singlet emitters with little distortion of the excited state. *Bodipy* dyes also exhibit exceptional stability,[5] making them attractive for diverse applications such as donor-acceptor systems,[6] artificial light harvesters,[7] fluorescent sensors,[8] laser dyes,[9] electron-transfer reagents[10] and light-emitting devices.[11]

To expand the utility of such dyes, we have developed recently an arsenal of synthetic methods by which the pseudo-meso 8-position,[12] the 2,6-pyrrolic position,[13] the 3,5-dimethyl fragments[14] and the 4,4′-boron positions can be routinely functionalized.[15] Alkyne substitution tools have been extensively applied in these procedures. By virtue of their inherent linearity and π-orbitals, triple bonds are not only an ideal conduit for electronic conductivity but also provide a facile entry into the construction of C–C-linked appendages.[16]

Thus, *Bodipys* are fascinating dyes, although serious drawbacks do exist which include the tendency to form aggregates and a very low solubility in polar solvents.[3] The few known examples of water-soluble *Bodipys* are all sulfonated species obtained either by direct reaction with chlorosulfonic acid at the 2,6 positions,[17] by Heck-type coupling[18] to introduce hydrophilic (sulfonated) substituents at the 2,6 positions, by reaction, again at the 2,6 positions, with chlorosulfonic acid after the introduction of other functionality[19] or by nucleophilic aromatic substitution of a 3-chloro substituent with 2-mercaptosulfonic acid.[20]

In this context, we report herein our attempts to prepare new water-soluble *Bodipy* dyes appropriately functionalized in several positions, together with description of the spectroscopic properties of the new dyes in various polar solvents. Two synthetic pathways were envisaged: (i) introduction of a protected sulfonate connected via a 'soft' coupling reaction to the dye or (ii) a post-synthetic introduction of a sulfobetaine unit. The critical aspect of this work concerns the use of sulfobetaines as zwitterionic templates to promote water solubility. This is the first application of such a procedure to *Bodipy* functionalization. Its initial step was the attachment of the protected sulfonates **1** to **4** to various *F-Bodipys*.[21]
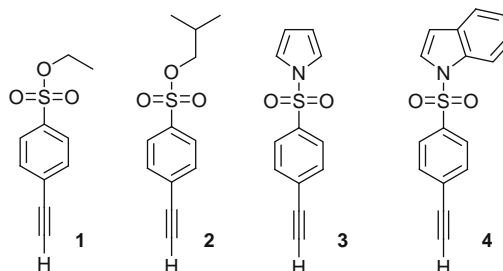
* Corresponding authors. Tel.: +33 3 90 24 26 89; fax: +33 (0) 3 90 24 27 61.
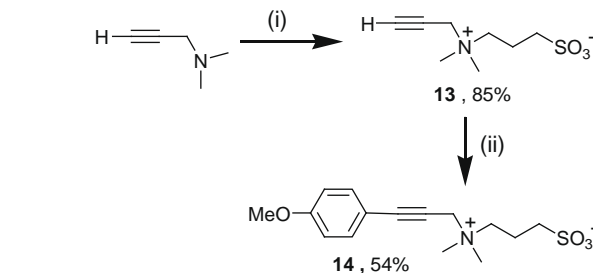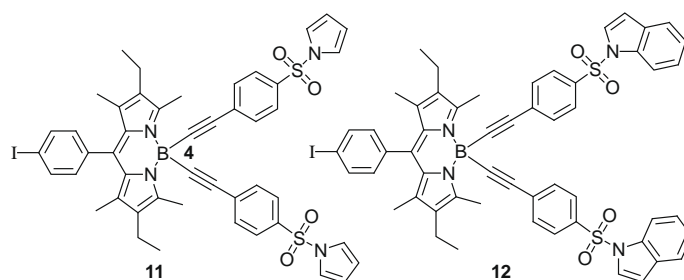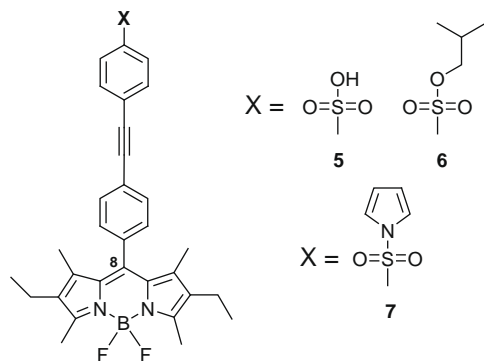*E-mail addresses:* gulrich@chimie.u-strasb.fr (G. Ulrich), ziessel@chimie.u-strasbg.fr (R. Ziessel).
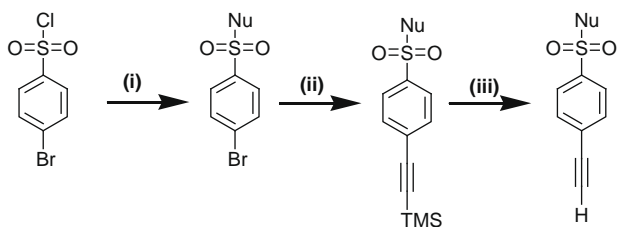
Scheme 1. Reagents and conditions: (i) isobutanol, 2,6-lutidine, 1,2-dichloroethane, rt, 70%; plus either pyrrole, NaH, THF, rt, 76%; or isoindole, NaH, THF, rt, 67%; (ii) TMSacetylene, TEA, benzene, [Pd(PPh$_3$)$_2$Cl$_2$] (0.06 mol %), CuI (0.1 mol %), 50 °C, 80% to 94%; (iii) KF, methanol, rt, 83–97%.

Scheme 2. Reagents and conditions: (i) 1,3-propanesultone, toluene, rt; (ii) 4-iodoanisole, [Pd(PPh$_3$)$_4$] (6 mol %), DMF, TEA, 80 °C.

These derivatives were prepared in three steps from 4-bromophenylsulfonyl chloride according to Scheme 1.
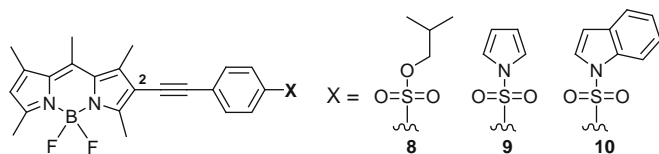
The cross-coupling reaction of **1**–**4** with dyes bearing halo-aromatic groups, such as 8-(4-iodophenyl)-*Bodipy*[12] is very efficient in the presence of TEA, benzene and [Pd(PPh$_3$)$_4$] (6 mol %) at 60 °C, leading to the desired compounds **5** to **7** in 85–91% yields.

Note that during the reaction with the terminal alkyne **1** the ethyl sulfoester is hydrolyzed. Following the same strategy, efficient cross-coupling with the 2-iodo-, or 2,6-diiodo-*Bodipy*[13] provided the substituted dyes **8**–**10** in excellent isolated yields (64–83%).

The pyrrole- or indole-protected sulfonates can be used to substitute F by acetylide at B by means of the corresponding Grignard reagents.[15] These were prepared from compounds **3** and **4** and EtMgBr in anhydrous THF at 60 °C. The resulting boron-functionalized dyes **11** and **12** were obtained in satisfactory yields after conventional purification procedures. Deprotection of the sulfonate, however, appeared to be difficult under various acidic or basic conditions. These are known to work well in other cases.[22]

As the basis for an alternative strategy, we examined the reactivity of the ethynylsulfobetaine derivative **13** towards Sonogashira cross-coupling reaction catalyzed by low valent Pd(0) (Scheme 2). Compound **13** was conveniently prepared from 1-(*N,N*-dimethylamino)-prop-2-yne and was found to react readily with 4-iodoanisole (as model compound) under classical cross-coupling reactions to afford the water-soluble compound **14**.
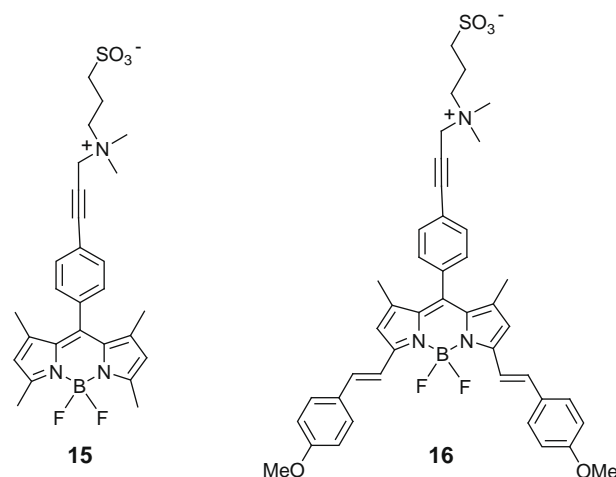
This encouraging result led us to examine the cross-coupling reaction of **13** with *Bodipy* dyes to see how greatly their water solubility might be enhanced. Dyes **15** and **16** were easily prepared from the corresponding iodophenyl derivatives,[14] and were obtained in 74 and 30% yields, respectively, after purification by flash chromatography on silica. The first two sulfobetaine-Bodipys are not highly soluble in water, unfortunately, tending to form aggregates as a result of their amphiphilic character. Thus, it appeared necessary to introduce hydrophilic substituents onto the aromatic core of the dyes.

With this objective, we turned our attention to cross-coupling reaction with *Bodipys* bearing a reactive iodo function at the pyrrolic ring (2 position). Surprisingly, the reaction did not produce the target betaine but produced a rather complicated mixture of highly coloured and non-fluorescent compounds that were difficult to separate properly. To circumvent this problem, we designed a two-step protocol based on (i) Pd(0) cross-coupling the dye with 1-dimethylamino-2-propyne and (ii) quaternization with 1,3-propanesultone under apolar conditions.[23] We first tested the concept

on a pyrene model compound. Both steps proved to be straightforward and compounds **17** and **18** were obtained in excellent yields, providing a water-soluble pyrene. Thus, we cross-coupled 1-dimethylamino-2-propyne with *Bodipy* **19**,[13] affording compound **20** in 82% yield, and subsequent alkylation with 1,3-propanesultone readily provided the desired water-soluble dye **21**. The use of a non-polar solvent caused the desired dye to precipitate during the course of the reaction and betaine **21** was isolated by centrifugation without additional treatment.
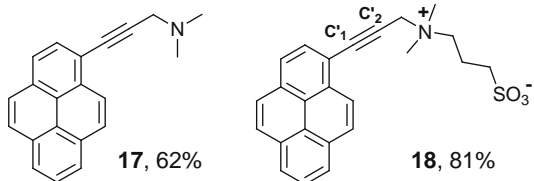
A single-crystal structure determination[24] for compound **18** established its expected structure (Fig. 1), with a C≡C distance of 1.185(4) Å and the puckered sulfobetaine chain folded towards the pyrene ring. The mean plane of the sulfobetaine chain lies close to that of the ethynylpyrene unit, the quaternary-N and sulfonate-S atoms lying approximately 1 Å to opposite sides of this plane. Within the lattice, the molecules form columnar stacks parallel to *a* (Fig. 2), with the pyrene substituent orientation alternating down the column. The normal to the pyrene plane is inclined at 20.5° to axis-*a* and alternates in orientation from one column to the next. While π-stacking is commonly observed in the solid state structures of pyrene and its derivatives,[25] and, in the present case, the rings in the columns down to axis-*a* are close to parallel (mean dihedral 2.5°) and separated by only 3.5 Å, consistent with such an interaction, analysis of the structure using Hirshfeld surfaces generated with CrystalExplorer[26] indicates that π-stacking is less important than interactions of the peripheral hydrogen atoms probably involving CH···O bonding (note that there is some evidence that the ethyne unit is involved in interaction with an adjacent pyrene ring). Thus, within a column, there are intermolecular contacts such that C16A is 3.502 Å from (sulfonate) O3, with C6A 3.596 Å from the same oxygen, while between columns, again
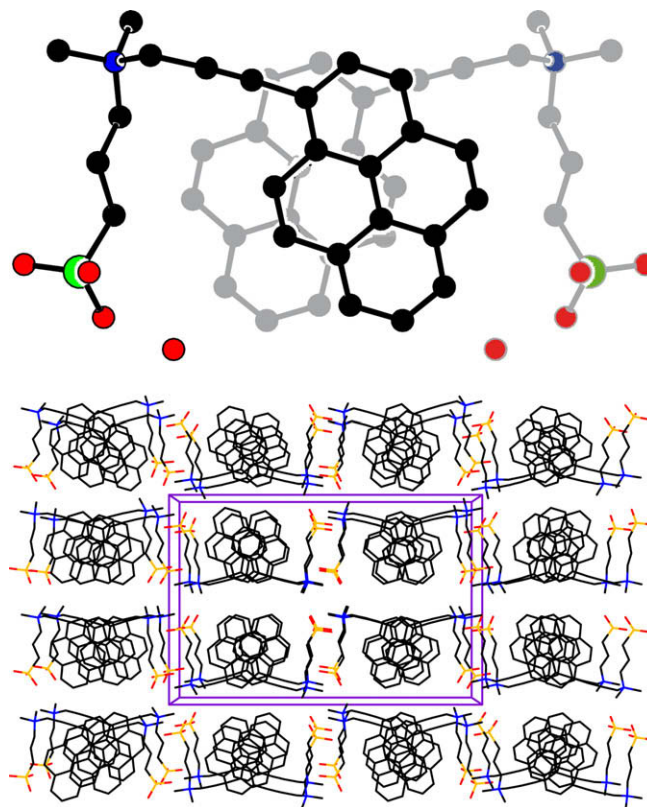

Figure 2. Top: Non-centrosymmetric π-stacked pairs viewed down the axis-*a*. In grey, molecule at positions $x - 1/2$, y, $1/2 - z$; hydrogen atoms are omitted for clarity. Bottom: Enlarged view of the crystal packing down the axis-*a*. The water molecules are omitted for clarity.

the same sulfonate-O has a contact to C5A of 3.502 Å. Note that there is also a short contact of O1W to C16A of 3.266 Å, though the (water) oxygen atom lies well out of the plane of the ring and thus the interaction may be of π···O character. This water molecule is clearly involved in H-bonding to sulfonate-O2 (O···O 2.803 Å) and has another short contact of 3.319 Å to C2 in an adjacent column. In any case, the Hirshfeld surface properties indicate that the strongest lattice interactions are those between the zwitterionic arms, which form head-to-tail arrays (i.e., antiparallel alignment of the dipoles) in columns also parallel to *a*. Interactions
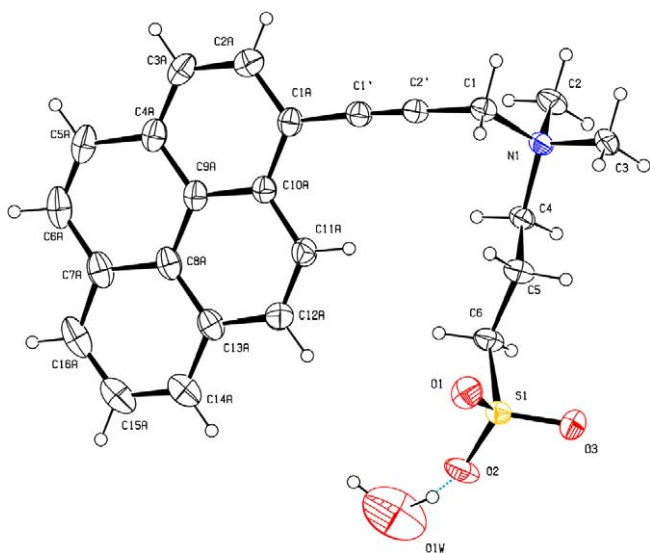

Figure 1. The molecular structure of **18**, showing the atom-labelling scheme. Displacement ellipsoids are drawn at the 30% probability level and H atoms are shown as small spheres of arbitrary radii.
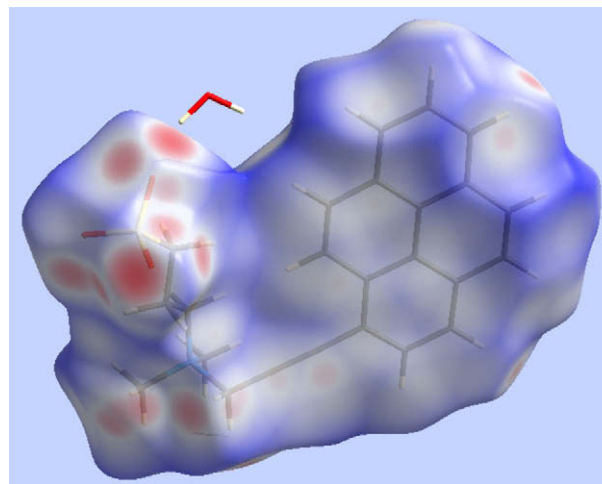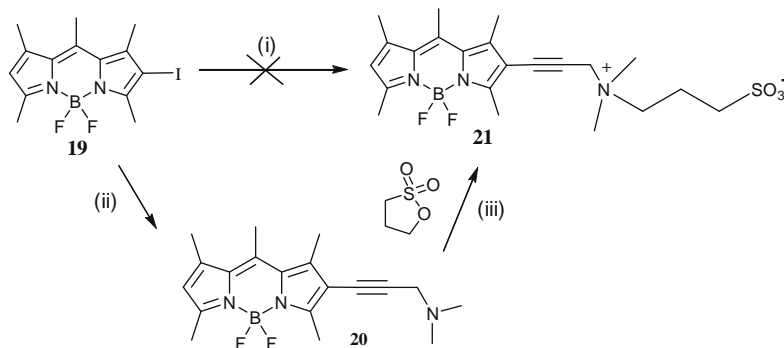
Figure 3. A view of the Hirshfeld surface calculated for the stoichiometric unit of crystalline compound **18**. By convention, red regions indicate points of unusually close contact to adjacent atoms.

Scheme 3. Reagents and conditions: (i) Compound **13**, [Pd(PPh₃)₄] (6 mol %), DMF, TEA, 80 °C; (ii) 1-dimethylamino-2-propyne, [Pd(PPh₃)₂Cl₂] (6 mol %), CuI (10 mol %), benzene, TEA, 60 °C, 82%; (iii) 1,3-propanesultone, toluene, 58 °C, 78%.
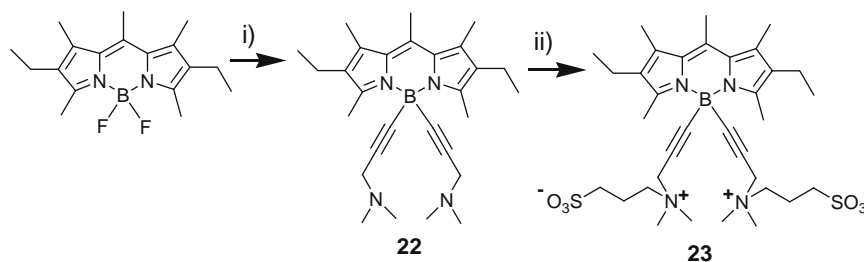
Scheme 4. Reagents and conditions: (i) 1-(N,N-dimethylamino)-propyne magnesium bromide, THF, 60 °C, 55%; (ii) propanesultone, toluene, 60 °C, 78%.
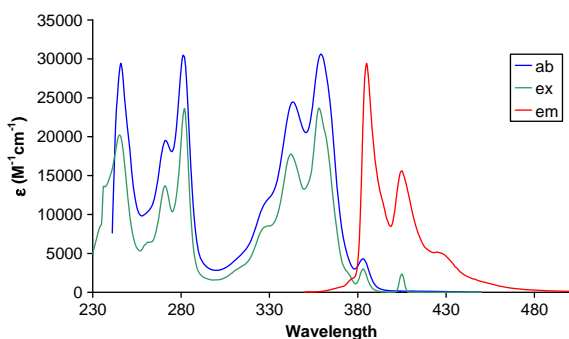

Figure 4. Absorption, emission and excitation spectra of **18** in water.

between the formally oppositely charged ammonium and sulfonate centres appear again to be mediated through CH···O interactions (Fig. 3).

Finally, this interesting strategy was applied towards the grafting of such betaine residues onto boron using the Grignard

analogue of 1-(N,N-dimethylamino)-prop-2-yne. The alkyne-substituted dye **22** was prepared in 55% isolated yield, then alkylated in toluene with 1,3-propanesultone as the reagent to give compound **23** in 78% yield after collection by centrifugation and washing with common apolar solvents (Schemes 3 and 4).

The electronic absorption spectrum of the water-soluble pyrene **18** exhibits characteristic π,π^* transitions for the various S_3, S_2 and S_1 excited states. Excitation spectra match the absorption spectra allowing to conclude that all the transitions are involved in the single and structured emission centred at 368 nm (Fig. 4). This emission mirrors the absorption and its single exponential decay in the nanosecond regime is typical of a singlet excited state. The water-soluble Bodipy **23** has a weak absorption at 368 nm typical of a $S_0 \rightarrow S_2$ transition and a strong absorption at 510 nm typical of a $S_0 \rightarrow S_1$ transition (Fig. 5). The short lifetime (9.2 ns) and the high quantum yield (81%) are in line with a singlet emitter. Finally, the blue dye **16** exhibits the characteristic structured absorption with a maximum at 643 nm and a pronounced intramolecular charge transfer (ICT) absorption at 369 nm (Fig. 6). The intense red emission at 660 nm has a short lifetime typical of a singlet
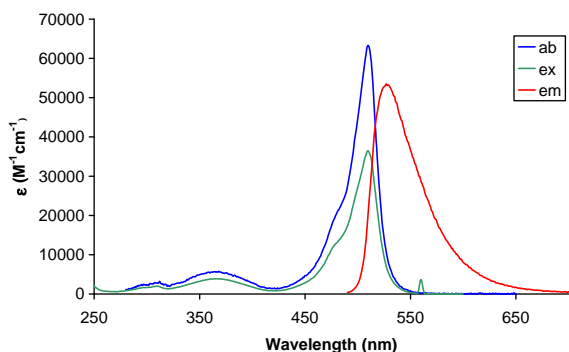

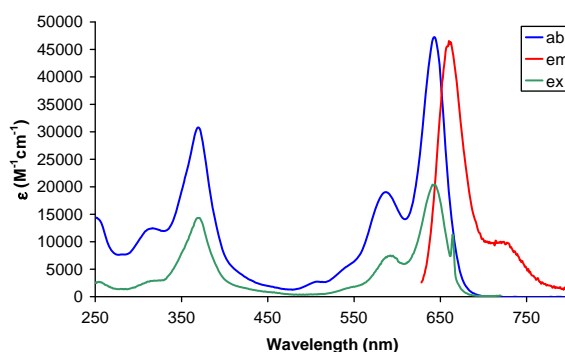**Figure 5.** Absorption, emission and excitation spectra of **23** in water.

Figure 6. Absorption, emission and excitation spectra of **16** in EtOH.

Table 1
Optical properties of selected compounds at rt[a]

Compds	λ_{abs} (nm)	ε_{max} (M^{-1} cm^{-1})	λ_{em} (nm)	τ_{em} (ns)	Φ_F (%)	k_r (10^8 s^{-1})	k_{nr} (10^6 s^{-1})
5 (EtOH)	524	54,100	537	4.3	66	1.53	79
6 (CH_2Cl_2)	527	65,000	542	5.1	48	0.94	102
7 (CH_2Cl_2)	527	92,600	541	5.5	64	1.16	65
8 (CH_2Cl_2)	523	67,000	548	4.5	78	1.73	49
9 (CH_2Cl_2)	523	48,000	548	3.9	79	2.04	54
10 (CH_2Cl_2)	523	71,500	548	5.1	74	1.47	49
11 (CH_2Cl_2)	522	103,000	534	6.9	50	0.72	72
12 (CH_2Cl_2)	523	54,000	534	6.4	82	1.61	35
15 (EtOH)	501	77,600	512	2.55	43	1.68	223
15 (H_2O)	496	75,500	511	3.18	20	0.63	252
16 (EtOH)	643	47,000	660	4.66	60	1.29	86
18 (H_2O)	359	30,600	358/ 405	11.6	32	0.28	59
18 (DMSO)	365	39,000	387/ 407	17.4	38	0.22	36
21 (DMSO)	506	44,700	529	7.2	80	1.11	28
23 (H_2O)	510	63,300	528	9.2	81	0.88	21

[a] Quantum yields determined in dilute solution ($c \sim 1.10{-}6$ M) using Rhodamine 6G as reference ($\Phi_F = 0.78$ in water, $\lambda_{exc} = 488$ nm) or quinine sulfate ($\Phi_F = 0.55$ in 1 M H_2SO_4, $\lambda_{exc} = 366$ nm).[27] All Φ_F are corrected for changes in refractive index. k_r and k_{nr} were calculated using the following equations: $k_r = \Phi_F/\tau_F$, $k_{nr} = (1 - \Phi_F)/\tau_F$, assuming that the emitting state is produced with unit quantum efficiency.

emission and a 60% quantum yield in ethanol. In fact, most of the radiative rate constants of around 10^8 s^{-1} are the same within experimental error (Table 1). However, the radiative rate constants are significantly weaker for the pyrene compounds **18** compared to those of the others, mostly because of the weaker quantum yield, which may be attributed to the presence of aggregates.

In short, we have established new synthetic routes for functionalization of *Bodipy* dyes with sulfonate residues. The use of unusual betaine building blocks carrying a terminal alkyne allows cross-coupling reactions to be effective under mild operational conditions. In some cases, a two-step synthesis is required to first graft the 1-dimethylamino-2-propyne fragment either on the dipyrromethene core or on the boron and to then quaternize the tertiary amine centre with 1,3-propanesultone. Interestingly, in all cases the optical properties are retained with respect to the non-substituted *Bodipys*. This work opens the door for the engineering of fluorescent dyes in aqueous medium and a vast range of new molecules, some being highly hydrophilic, can now be envisaged. By extension of these protocols, the engineering of amphoteric dyes bearing large functionalities and which are potentially able to be properly grafted on surfaces should also be possible. Work along these lines is currently in progress.

References and notes

1. Mishra, A.; Behera, R. K.; Behera, P. K.; Mishra, B. K.; Behera, G. B. *Chem. Rev.* **2000**, *100*, 1973–2012.
2. Ziessel, R.; Ulrich, G.; Harriman, A. *New J. Chem.* **2007**, *31*, 496–501.
3. Loudet, A.; Burgess, K. *Chem. Rev.* **2007**, *107*, 4891–4932.
4. (a) Ziessel, R. *Compt. Rendus Acad. Sci. Chim.* **2007**, *10*, 622–629; (b) Ulrich, G.; Ziessel, R.; Harriman, A. *Angew. Chem., Int. Ed.* **2008**, *47*, 1184–1201.
5. Haughland, R. P. *Handbook of Fluorescent Probes and Research Products*, 9th ed.; Molecular Probes: Eugene, OR, 2002.
6. Coskun, A.; Akkaya, E. U. *J. Am. Chem. Soc.* **2006**, *128*, 14474–14475.
7. Harriman, A.; Izzet, G.; Ziessel, R. *J. Am. Chem. Soc.* **2006**, *128*, 10868–10875.
8. Bricks, J. L.; Kovalchuk, A.; Trieflinger, C.; Nofz, M.; Büschel, M.; Tolmachev, A. I.; Daub, J.; Rurack, K. *J. Am. Chem. Soc.* **2005**, *127*, 13522–13529.
9. Lopez-Arbeloa, F.; Banuelos Prieto, J.; Martinez Martinez, V.; Arbeloa Lopez, T.; Lopez Arbeloa, I. *Chem. Phys. Chem.* **2004**, *5*, 1762–1771.
10. Sartin, M. M.; Camerel, F.; Ziessel, R.; Bard, A. J. *J. Phys. Chem. C* **2008**, *112*, 10833–10841.
11. Lai, R. Y.; Bard, A. J. *J. Phys. Chem. B* **2003**, *107*, 5036–5042; Hepp, A.; Ulrich, G.; Schmechel, R.; von Seggern, H.; Ziessel, R. *Synth. Metals* **2004**, *146*, 11–15.
12. Ulrich, G.; Ziessel, R. *J. Org. Chem.* **2004**, *69*, 2070.
13. Bonardi, L.; Ulrich, G.; Ziessel, R. *Org. Lett.* **2008**, *10*, 2183–2186.
14. Ziessel, R.; Ulrich, G.; Harriman, A.; Alamiry, M. A. H.; Stewart, B.; Retailleau, P. *Chem. Eur. J.* **2009**, *15*, 1359–1369.
15. Goze, C.; Ulrich, G.; Ziessel, R. *J. Org. Chem.* **2007**, *72*, 313–322; Goze, C.; Ulrich, G.; Ziessel, R. *Org. Lett.* **2006**, *8*, 4445–4448; Ziessel, R.; Goze, C.; Ulrich, G. *Synthesis* **2007**, 936–949.
16. 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.
17. Boyer, J. H.; Haag, A. M.; Sathyamoorthi, G.; Soong, M. L.; Thangaraj, K. *Heteroatom Chem.* **1993**, *4*, 39–49.
18. Thivierge, C.; Bandichhor, R.; Burgess, K. *Org. Lett.* **2007**, *9*, 2135–2138.
19. Li, L.; Han, J.; Nguyen, B.; Burgess, K. *J. Org. Chem.* **2008**, *73*, 1963–1970.
20. Li, L.; Nguyen, B.; Burgess, K. *Bioorg. Med. Chem. Lett.* **2008**, *18*, 3112–3116.
21. Roberts, J. C.; Gao, H.; Gopalsamy, A.; Kongsjahju, A.; Patch, R. J. *Tetrahedron Lett.* **1997**, *38*, 355–358.
22. Jolicoeur, B.; Chapman, E. E.; Thompson, A.; Lubell, W. D. *Tetrahedron* **2006**, *62*, 11531–11563.
23. Kong, X.; Migneault, D.; Valade, I.; Wu, X.; Gervais, F. US Patent 2007/0010573 A1, 2007.
24. *The crystal data of* **18**: $C_{24}H_{23}NO_3$, H_2O, $M = 423.51$, orthorhombic, space group *Pbca*, $a = 7.517(2)$, $b = 19.331(3)$, $c = 28.791(4)$ Å, $V = 4183.7(14)$ Å^3, $Z = 8$, $D_c = 1.345$ g cm^{-3}, μ(Mo-Kα) = 0.186 mm^{-1}, $F(0\,0\,0) = 1792$, prismatic crystal, yellow, size = $0.60 \times 0.30 \times 0.10$ mm, 21,734 reflections measured [$R_{int} = 0.047$], 3834 unique, w$R_2 = 0.141$ for all data, conventional $R = 0.051$ [$(\Delta\sigma)_{max} = 0.001$] on *F*-values of 2404 reflections with $I > 2\sigma(I)$, $S = 1.018$ for all data and 273 parameters. Unit cell determination and intensity data collection ($2\theta = 50.7°$) were performed on a Enraf-Nonius KAPPA CCD diffractometer at 293(2) K. Structure solution by direct methods and refinement by full-matrix least-squares on P^2. Programs: COLLECT [Nonius, B. V. (1999)], HKL2000 [Otwinowski, Z.; Minor, W. 1997], SHELX97 [Sheldrick, G. M. 2008]. CCDC [deposit No: 716693] contains the supplementary crystallographic data for this Letter. These data can be obtained free of charge from the Cambridge Crystallographic Data Centre via www.ccdc.cam.ac.uk/data_request/cif.
25. (a) Robertson, J. M.; White, J. G. *J. Chem. Soc.* **1947**, 358–368; (b) Ziessel, R.; Goze, C.; Ulrich, G.; Césario, M.; Retailleau, P.; Harriman, A.; Rostron, J. P. *Chem. Eur. J.* **2005**, *11*, 7366–7378.
26. Spackman, M. A.; Jayatilaka, D. *CrystEngCommun* **2009**, *11*, 19–32.
27. Olmsted, J., III *J. Phys. Chem.* **1979**, *83*, 2581–2584.