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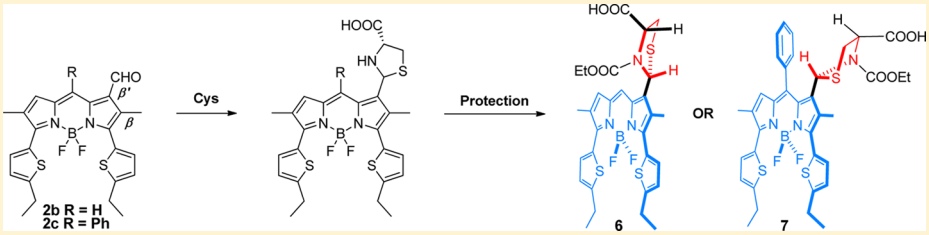
Thiazolidine Derivatives from Fluorescent Dithienyl-BODIPY-carboxaldehydes and Cysteine

Arnaud Poirel, Antoinette De Nicola,* and Raymond Ziessel*

Institut de Chimie et procédés pour l'Energie, l'Environnement et la Santé (ICPEES), Laboratoire de Chimie Organique et Spectroscopies Avancées (LCOSA), UMR 7515, École de Chimie, Polymères, Matériaux de Strasbourg (ECPM), 25 Rue Becquerel, 67087 Strasbourg Cedex 02, France

Supporting Information



ABSTRACT: Fluorescent dithienyl-borondipyrromethene (BODIPY) dyes formylated in the β'-position (**2b**, **2c**) have been treated with L-cysteine to provide thiazolidine derivatives. N-Protection of the thiazolidine unit by ethoxycarbonylation facilitated isolation of the two major diasteroisomers **6** and **7**. These stereoisomers have been fully characterized by ^{1}H NMR spectroscopy, allowing assignment of their stereochemistry as 2*R*,4*R*,a*S* and 2*S*,4*R*,a*R*, respectively. The optical properties of the thiazolidine dyes differ markedly in both absorption (λ_{abs} = 612 nm for **6** and 615 nm for **7**) and emission (λ_{em} = 669 nm, Φ_F = 62% for **6** and λ_{em} = 672 nm, Φ_F = 19% for **7**) from those of the BODIPY-carboxaldehydes **2b** (λ_{abs} = 643 nm and λ_{em} = 719 nm, Φ_F = 26%) and **2c** (λ_{abs} = 636 nm and λ_{em} = 710 nm, Φ_F = 36%). In a mixed solvent [phosphate buffer saline (PBS), pH = 7.4/ethanol 1:9], the fluorescence response of the dyes in the presence of L-cysteine is slow, but a ratiometric detection process in the therapeutic window (650 to 800 nm) is evident.

INTRODUCTION

The thiazolidines are saturated five-membered-ring heterocycles where one S and one N atom are present in a 1,3 array. These heteroatoms can be associated with biological activity in many thiazolidines.[1] Natural products[2] containing this ring have been isolated and characterized, the best known being penicillin. Over the past decades, a plethora of synthetic analogues have been scrutinized for their specific pharmaceutical activities. They have proved to have interesting antimicrobial, anti-inflammatory, anticonvulsant, antimalarial, analgesic, anti-HIV, and anticancer activity.[2,3] In fact, many thiazolidines exist in equilibrium with the open chain Schiff-base[4] and its carbonyl and aminothiol precursors.[5] One straightforward way to obtain substituted thiazolidines is to treat α-aminothiols with carbonyl derivatives. The presence of a substituent on the nitrogen atom stabilizes the thiazolidine ring[6] and prevents (i) decomposition to either the imine or the precursors and (ii) C(2)-epimerization. Work concerning the steric demand of the substituent during the condensation reactions between an aldehyde and an aminothiol has analyzed effects on the C(2)-stereochemistry.[7] Usually, the formation of 2-substituted thiazolidine-4-carboxylic acids from cysteine (Cys) provides a mixture of stereoisomers. Several examples from the literature show that this reaction exhibits stereoselectivity generally when, besides the cysteine, the aldehyde is also chiral.

When the aldehyde used is a fluorophore [such as a boron dipyrromethene (BODIPY)[8,9] or a diketopyrrolopyrrole,[10] a cyanine[11] or a squaraine[12]], the condensation reaction with Cys can be considered as a detection method for this amino acid.[13] A deficiency of Cys in vivo is involved in many syndromes such as hair depigmentation, liver damage, and physical fatigue[14] whereas elevated Cys in human blood is a risk factor for Alzheimer's and cardiovascular diseases as well as for osteoporosis.[15] Due to the crucial role played by thia-biomolecules and thiols in biological systems the development of specific fluorogenic probes is an expanding research subject.[16,17] Several detection processes have been engineered involving Michael addition,[18] cleavage of sulfonamide and sulfonate esters,[19] cleavage of the selenium−nitrogen bond and of disulfides,[20] metal complex redox processes,[21] and metal complex replacement of ligands.[22] Recently, the use of nanoparticles (or nanorods) of gold,[23] of silver (clusters),[24] and of fluorescent organic particles (FONs) have been studied.[25] The cyclization reaction of an aldehyde leading to a thiazolidine[26] engaged our attention due to the facts that (i) fluorogenic aldehydes are relatively easy to produce, (ii) spectral changes in the reaction can be dramatic due to the loss of the aldehyde chromophore, (iii) steric demand around the

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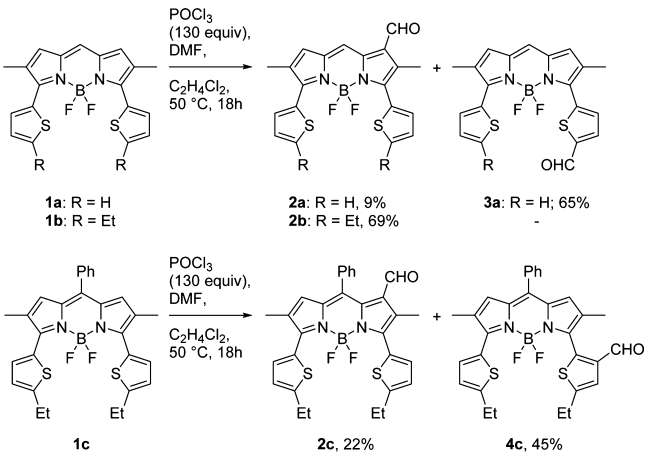
aldehyde function is easily tailored during the construction of the dye, and finally (iv) the solubility and detection window can be tuned by chemical engineering. Of the various available and appealing fluorophores, the NIR emitters of the borondipyrromethene (BODIPY) family are particularly interesting.[27,28] Indeed any system with an optical response in the NIR region is ideal for therapeutic applications because the interference and autofluorescence from the endogenous chromophores are weak and the penetration of the tissues can be deeper.[29]

In this work a series of dithienyl-BODIPY carboxaldehydes has been synthesized. The two dyes **2b** and **2c** with aldehyde groups in the β'-position provided thiazolidine-4-carboxylic acid derivatives when reacted with L-cysteine. Their transformation into carbamates **6** and **7** stabilized the rings and enabled complete characterization of these derivatives, including specification of their absolute configuration by 2D ^{1}H NMR COSY and NOESY experiments. The formation of the thiazolidine ring has a characteristic optical response in absorption and steady-state fluorescence.

RESULTS AND DISCUSSION

Synthesis and Characterization. Three dithienyl-BODIPYs **1a**–**c** were used as starting materials. All of them were prepared by condensation of 2-thienylpyrroles with ethyl orthoesters [RC(OEt)$_3$]. The syntheses of **1a** and **1b** were carried out as described previously.[30] Dye **1c** differs from the others in having a phenyl group on the meso-position. Their formylation (Scheme 1) was achieved by a Vilsmeier–Haack

Scheme 1. Formylation of BODIPYs



reaction under conditions described by Jiao[31] for the synthesis of BODIPY-β-carboxaldehydes, and average yields of 70% were routinely obtained. Three sites of formylation were observed, with a regioselectivity depending on the substitution pattern of the BODIPY core.

In all cases, BODIPY-β'-carboxaldehydes were obtained, but the best result was achieved with **1b**. Here the most nucleophilic α-position of the thiophene ring is blocked by an ethyl group, and there is no steric hindrance in the meso-position. Thus, compound **2b** was the only aldehyde isolated in 69% yield. The BODIPY **1a** gave as the major product **3a** (65%) with a formyl group introduced at the free α-position of the thiophene ring, while the phenyl group in **1c** hindered the aromatic electrophilic substitution in the β'-position. Thus, two aldehydes were obtained from **1c**: a major product **4c** (45%) functionalized on the thiophene ring and the suited BODIPY-β'-carboxaldehyde **2c** (22%).

The position of the formyl group in **4c** was established unambiguously from both COSY and NOESY measurements (Figure 1). The most important aspect of the NOESY spectrum was the clear interaction revealed between the aldehydic proton and the methyl group of the adjacent pyrrole unit.

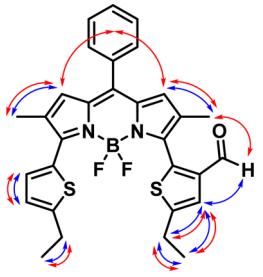
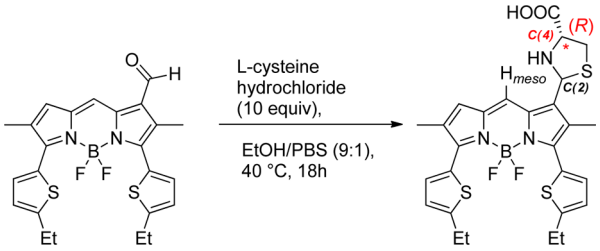


Figure 1. COSY (blue line) and NOESY (red line) correlations in compound **4c**.

The cyclocondensation reaction (Scheme 2) between **2b** and the HCl salt of L-cysteine was carried out at 40 °C for 18 h in a

Scheme 2. Thiazolidine Formation in PBS (pH = 7.4/EtOH)[a]



[a] H_{meso} accounts for the proton in the meso position. All other protons are not shown for the sake of clarity.

mixture of PBS (pH = 7.4) and EtOH (1:9, v/v) to give **5** in 55% yield after recrystallization. It is noteworthy that without the buffer the major product obtained is the highly fluorescent diethyl acetal of the aldehyde **2b** which might give false signals during the optical measurements. The ^{1}H NMR spectrum of **5** (Figure 2b) shows two sets of signals for nearly every proton, consistent with the product being a 1:1 mixture of two diastereomers. The most affected by the difference in stereochemistry are the protons H(4) (with $\Delta\delta$ = 0.49 ppm), H(2) (with $\Delta\delta$ = 0.07 ppm), and H_{meso} (with $\Delta\delta$ = 0.08 ppm). The mild conditions of the reaction exclude the epimerization of the atom of carbon C(4) (usually only observed at 100 °C with a mixed anhydride-type intermediate)[7b] of the thiazolidine ring. Thus, the stereoisomers obtained are considered to be C(2) epimers.

To separate the stereoisomers by column chromatography, the thiazolidine **5** was stabilized by the introduction of an ethoxycarbonyl substituent on the ring nitrogen N(3) (Scheme 3). The one-pot conditions from **2b** to obtain **6** have been optimized. After 20 h for the cyclocondensation reaction, the crude compound was washed with water to eliminate the salts and treated with ethyl chloroformate in THF for 1 h at room temperature. The purification by silica gel column chromatog-

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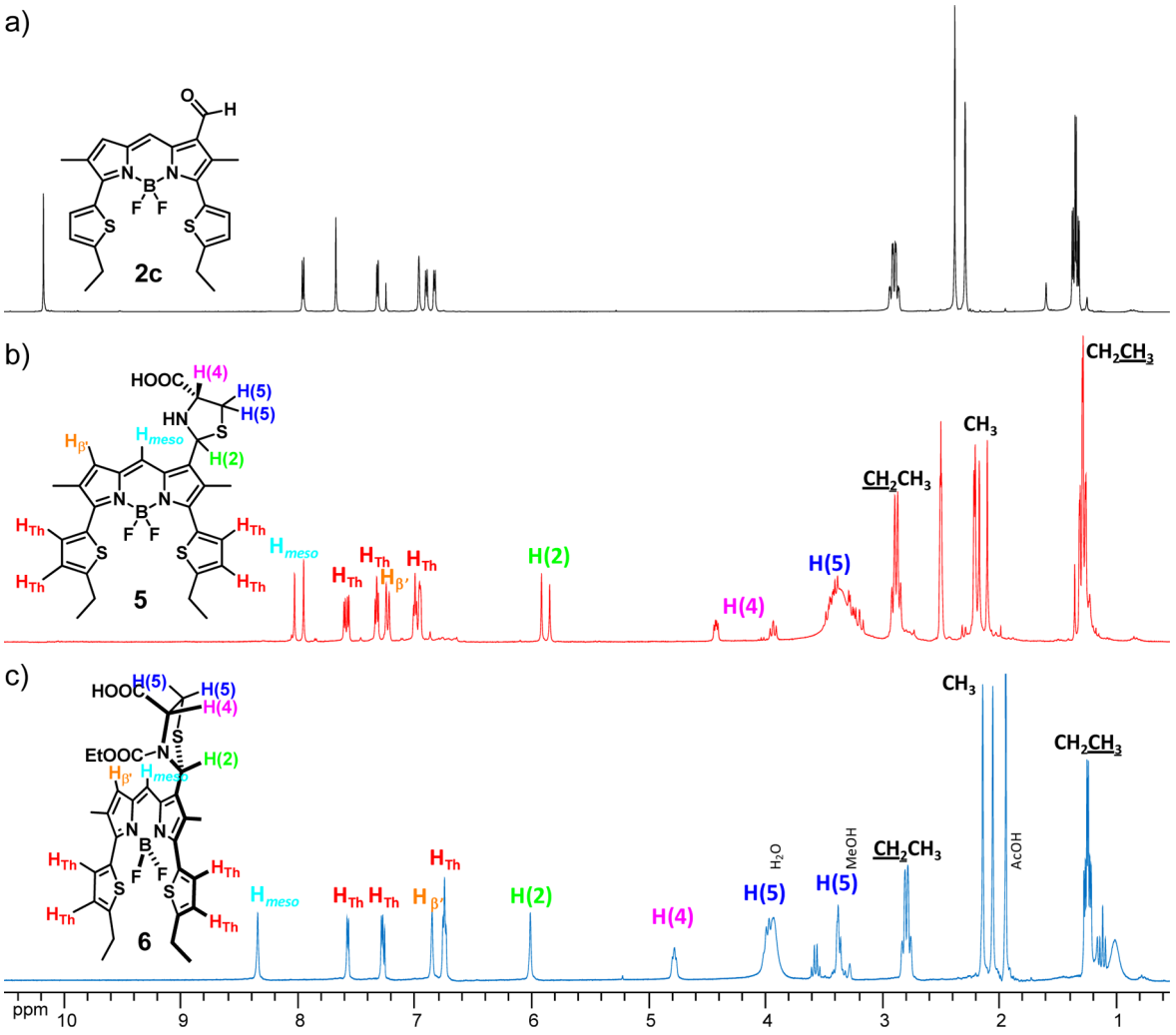
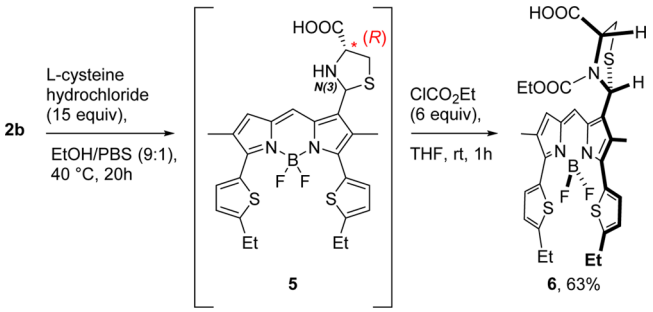


Figure 2. ^{1}H NMR spectra of compounds (a) **2c** in $CDCl_3$, (b) **5** in DMSO-d_6, and (c) **6** in $CDCl_3$.

Scheme 3. Protection of Thiazolidine



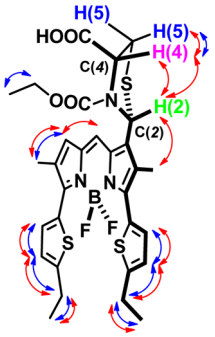


Figure 3. COSY (blue line) and NOESY (red line) correlations in compound **6**.

raphy gave two fractions. Their ^{1}H NMR spectra showed that the major product (63%) was the single diastereoisomer **6** (Figure 2c) and the minor one (20%) a mixture of two other diastereoisomers. The nature of the minor products was not established but spectral data [IR, NMR (^{1}H, ^{11}B, ^{13}C, COSY, NOESY, MS)] and elemental analyses were consistent with the structure and the absolute configuration assigned to **6**.

The NOESY correlations (Figure 3) for compound **6** brought to light the fact that H(2) and H(4) project from the same face of the thiazoline ring and that H(2) interacts with the methyl group in the *β*-position of the BODIPY but not at all with the proton H_m (labeled in Scheme 2). This indicates that there is a hindered rotation about the single bound connecting the thiazolidine ring and the BODIPY moiety. This is easily evidenced by CPK (Corey, Pauling, Kolton) molecular models. Thus, this atropoisomer **6** displays three chiral elements: two chiral centers (C(2) and C(4) of the thiazolidine) and one chiral axis; its stereochemistry is assigned as 2*R*,4*R*,a*S*, assuming that the configuration of the stereogenic C(4) of cysteine is retained in the reaction. Unfortunately the measurement of the specific rotary power failed because of the deep blue color of the solution (even at $c = 5 \times 10^{-6}$ M).

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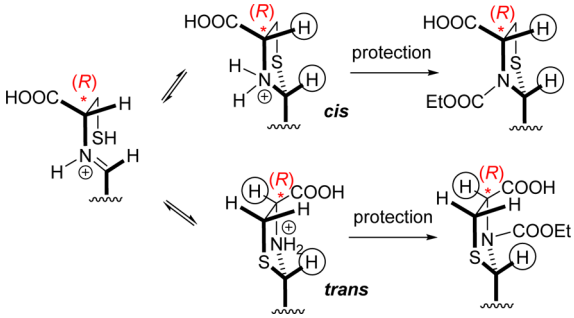
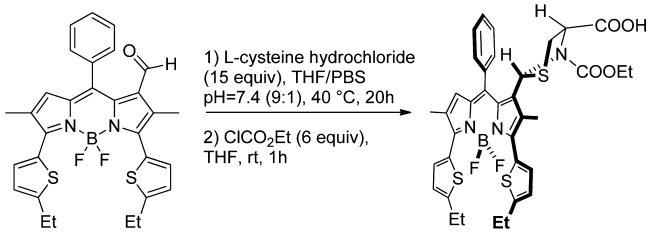
Scheme 4. Proposed Ring-Opening Mechanism of Thiazolidine 5 Protection



Scheme 5. Protection of Thiazolidine



Surprisingly, although the cyclocondensation products of **2b** with the L-cysteine consisted of a 1:1 mixture of C(2) epimeric thiazolidine-4-carboxylic acids **5**, the protection of this mixture (checked to still be 1:1 immediately before the treatment with ethyl chloroformate) resulted in the isolation of **6** (a *cis*-thiazolidine stereoisomer) as the major product. This can be explained if N-protection is accompanied by partial inversion at C(2). This inversion could take place through a ring-opening mechanism involving the Schiff-base intermediate in equilibrium with the protonated cis and trans forms (Scheme 4). The preferential formation of the cis stereoisomer remains unclear but can be rationalized by assuming that its protection step is faster than for the trans stereoisomer and that the cis stereoisomer is the kinetic product of the reaction. The steric congestion for the cis and trans isomers are similar, and no specific intramolecular interaction for both forms could be envisaged at this stage.

The same one-pot reaction was conducted with the aldehyde **2c** (Scheme 5). The low solubility of this compound in EtOH led us to use saline phosphate buffer (PBS) at pH = 7.4/THF (1:9, v/v) as a solvent mixture for the cycloaddition step with the L-cysteine reagent. The protection step with 6 equiv of ethyl chloroformate resulted in the formation of a single diastereoisomer **7** in 40% yield. In parallel, 47% of the starting material **2c** was recovered and recycled.

Spectral data [IR, NMR (^{1}H (Figure 4), ^{11}B, ^{13}C, COSY, NOESY (Figure 5)), MS] and elemental analyses for **7** were consistent with the structure shown in Scheme 5.

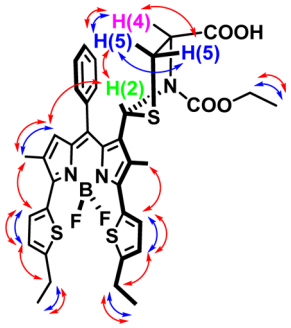


Figure 5. COSY (blue line) and NOESY (red line) correlations in compound **7**.

As for compound **6**, this N-protected thiazolidine **7** has two chiral centers and one chiral axis. The NOESY experiment allows assignment of its absolute configuration. It shows interactions between H(2) and H(5) and between the same H(5) and H(4), meaning that these three protons are on the same face of the thiazolidine ring.

Again, this one-pot reaction results in the selective formation of the *cis*-thiazolidine stereoisomer. The NOESY correlations between H(2) and the phenyl protons (absent between H(2) and the methyl protons) showed as well that the substituents on the thiazolidine ring are oriented away from the phenyl group.

This structure is supported by ^{1}H NMR spectroscopy (Figure 4); the chemical shifts of protons H(2), H(4), H(5), and $H_{\beta'}$ (Table 1) are upfield compared to those of **6**. These protons are located in the shielding cone that results from the phenyl-moiety-induced magnetic field. Thus, stereochemistry of **7** has been assigned as 2*S*,4*R*,a*R*.

Optical Properties. Spectroscopic data relevant to the present discussion are collected in Table 2, and typical spectra are given in Figures S32 to S49 (Supporting Information) for

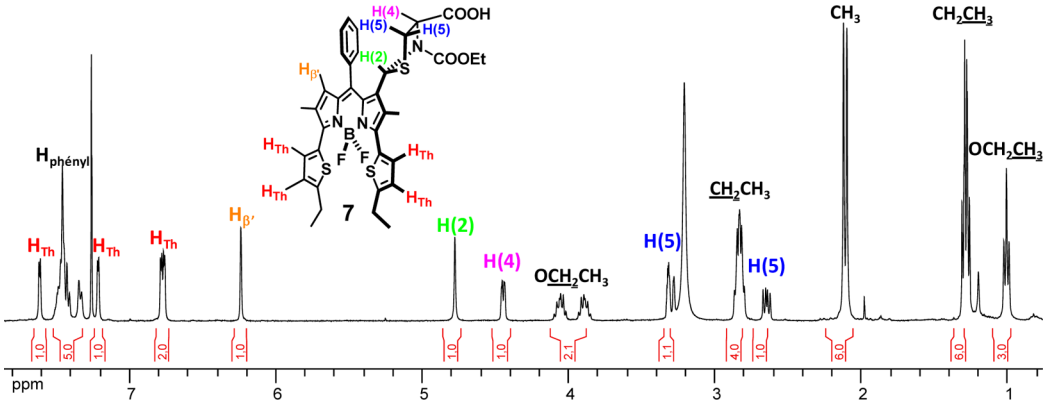


Figure 4. ^{1}H NMR spectrum of compound **7** (in $CDCl_3$).

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Table 1. Protons Chemical Shift (in $CDCl_3$) of Compounds 6 and 7

	δ (ppm)			
protons	Hβ′	H(2)	H(4)	H(5)
6	6.85	6.02	4.78	3.39
7	6.24	4.77	4.44	3.28/2.64

dyes **1c**, **2a**−**c**, **3a**, **4c**, **6**, and 7 in different solvents. The salient absorption spectra of the dithienyl starting materials **1a**−**c** show a strong $S_0 \rightarrow S_1$ transition (of π−π^* nature) in the 597 to 616 nm range (Figure 6 and Table 2).

This absorption is typical of BODIPY dyes and reflects the rigidity of the central core due to boron complexation.[32] The bathochromic shift observed for **1b** and **1c** reflects the electronic density increase imported by the ethyl group substituent on the thiophene rings. The presence of the phenyl residue in the meso-position of dye **1c** does not result in an increase of conjugation, explaining the unchanged value of λ_{abs}. Furthermore, the radiative and nonradiative deactivation rates remain similar, indicating that the phenyl ring is twisted out of the dipyrromethene plane and does not participate in the optical pathway. The quantum yields decrease slightly with an increase in the number of substituents, being 78% for **1a**, 69% for **1b**, and 62% for **1c**, while the singlet state lifetimes remain similar (8−10 ns).

The absorption properties of the aldehyde derivatives **2a**, **2b**, and **2c** (Figure 7a) are sensitive to the substitution pattern on the Bodipy core but also on the thienyl side rings. These compounds exhibit bathochromically shifted absorption maxima at respectively 624 nm (for **2a**), 643 nm (for **2b**), and 636 nm (for **2c**, Figure 7a), compared to those of the parent BODIPYs **1a** (597 nm), **1b** (616 nm), and **1c** (616 nm) because of the increase of π-conjugation in the BODIPY

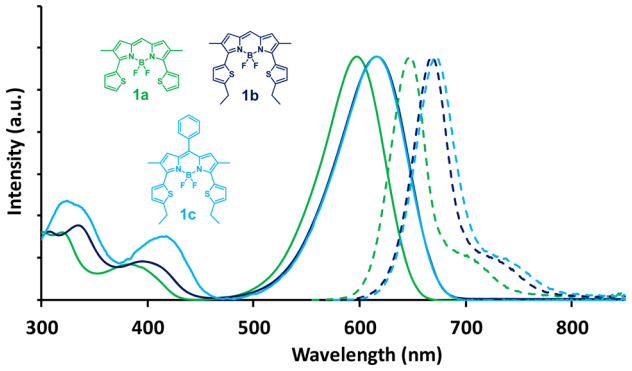

Figure 6. Absorption (plain trace) and emission (dashed trace) for BODIPY dyes **1a** (green), **1b** (dark blue), and **1c** (sky blue). All spectra were recorded in dichloromethane at 25 °C with c = 0.9 to 1.6 × 10^{-5} M for the absorption and c = 0.9 to 1.6 × 10^{-6} M for the emission.

frameworks. However, in the case of **2c** the *meso*-phenyl group prevents the carbonyl group being coplanar with the main core of the dye. Thus, the π-conjugation in **2c** is less pronounced compared to **2b** and a hypsochromic shift of 7 nm is observed.

Surprisingly, when the formyl group is located onto the thiophene ring, the electronic effects are weakly reflected in λ_{abs} unlike those of ethyl groups (Figure 7b). For **3a**, λ_{abs} at 604 nm is only slightly shifted compared to **1a** (λ_{abs} at 597 nm) and **4c** (λ_{abs} at 595 nm). For the latter case, the hypsochromic shift in the absorption (by 12 nm) is significant compared to the parent BODIPY **1c** (λ_{abs} = 616 nm). The position of the formyl group in **4c** causes an increase of dihedral angles between the thiophene ring and the core of the BODIPY and consequently decreases the π-conjugation in the molecule. As observed in general,[34] the aldehyde quantum yields are lower (Φ_F = 22−38%), with respect to the unsubstituted compounds **1a**−**c** (Φ_F

Table 2. Spectroscopic Data of Compounds 1 to 7 Determined at 25 °C

dye	λ_{abs}[a] (nm)	ε (M^{-1} cm^{-1})	λ_{em}[a] (nm)	Φ_F[b]	τ_F[c] (ns)	k_r[d] (10^8 s^{-1})	k_{nr}[d] (10^8 s^{-1})	Δ_s[e] (cm^{-1})	solvent
1a	606	58000	649	0.81	9.7	0.8	0.2	1100	toluene
	597	58000	647	0.78	9.2	0.8	0.2	1300	CH_2Cl_2
2a	624	49000	690	0.38	7.8	0.5	0.8	1500	CH_2Cl_2
3a	612	54000	666	0.67	–	–	–	1300	toluene
	604	51000	663	0.63	–	–	–	1500	CH_2Cl_2
	600	52000	656	0.55	–	–	–	1400	EtOH
	604	29000	664	0.10	–	–	–	1500	DMSO
1b	616	58000	668	0.69	10.3	0.7	0.3	1300	CH_2Cl_2
2b	651	54000	717	0.26	–	–	–	1400	toluene
	643	50000	719	0.22	–	–	–	1600	CH_2Cl_2
	636	50000	716	0.16	–	–	–	1800	EtOH
	639	19000	718	0.34	–	–	–	1700	DMSO
1c	616	54000	670	0.62	8.4	0.7	0.5	1300	CH_2Cl_2
2c	636	50000	710	0.36	4.6	0.8	1.4	1600	CH_2Cl_2
	632	52000	703	0.32	–	–	–	1600	EtOH
4c	595	53000	646	0.38	3.5	1.1	1.8	1300	CH_2Cl_2
6	612	45000	669	0.62	7.9	0.8	0.5	1400	CH_2Cl_2
	596	49000	650	0.96	–	–	–	1400	EtOH
7	615	47000	672	0.19	4.3	0.4	1.9	1400	CH_2Cl_2
	610	48000	665	0.30	–	–	–	1400	EtOH
	616	45000	673	0.22	–	–	–	1400	DMSO

[a]Error ±2 nm. [b]Tetramethoxy-BODIPY, Φ_F = 0.49 in dichloromethane (λ_{exc} = 650 nm) was used as reference.[33] All Φ_F were corrected with the refractive index of the medium, error ±10%. [c]Error ±0.5 ns. [d]Calculated using the following equations: $k_r = \Phi_F/\tau_F$ and $k_{nr} = (1-\Phi_F)/\tau_F$, assuming that the emitting state is produced with unit quantum efficiency. [e]Stokes shift: $\Delta s = (1/\lambda_{abs}) - (1/\lambda_{em})$.

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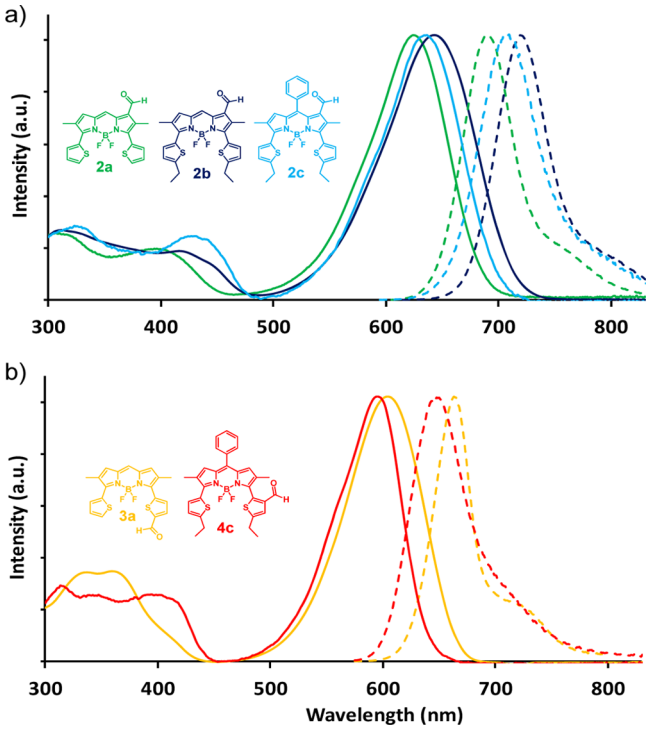

Figure 7. Absorption (plain trace) and emission (dashed trace) for BODIPY-carboxaldehydes: (a) **2a** (green), **2b** (dark blue), **2c** (sky blue); (b) **3a** (yellow) and **4c** (red). All spectra were recorded in dichloromethane at 25 °C with c = 1.1 to 1.8 × 10^{-5} M for the absorption and c = 1.1 to 1.8 × 10^{-6} M for the emission.

= 78−62%). No such decrease is observed for **3a** (Φ_F = 63%), highlighting the fact that the aldehyde in this substitution position does not contribute to the optical transition.

The less intense but more energetic optical transitions found in the 300 to 450 nm range are assigned to $\pi-\pi^*$ excitations of the thiophene subunits as well as to the n → π^* transitions of the aldehyde functions and the $S_0 \rightarrow S_2$ transition (of $\pi-\pi^*$ nature) of the BODIPY core.[35] Note that these transitions are sensitive to the degree and nature of the substitution and that the substitution position of the aldehyde α or β (**3a** or **4c**) on the thiophene ring greatly influences the energy of the $\pi-\pi^*$ transitions (Figure 7). The $\pi-\pi^*$ optical transition of the phenyl ring occurs below 300 nm.

The emission wavelengths are independent of the excitation wavelength, and in all cases the excitation spectra match the absorption spectra. This is in keeping with a singlet emission and the absence of aggregation under the conditions used. These dyes exhibit unusually large Stokes shifts of around 1400 cm^{-1} and have similar radiative and nonradiative deactivation rates constants (Table 2).

The isolated N-protected thiazolidines **6** and **7** show absorption and emission profiles essentially the same as the parent BODIPYs in CH_2Cl_2 except for their quantum yields (Table 2). For **6** it is the same in CH_2Cl_2 (62%) but remarkably higher in EtOH (96%). For **7** the quantum yield decreases to 19% in CH_2Cl_2. It is noteworthy that in EtOH its value is higher (30%) but not as good as for **6**. These values reflect the fact that the BODIPY core in **7** is probably more buckled compared to **6** due to the steric demand of the phenyl and thiazolidine moieties. In both cases the excitation spectra match the absorption spectra over the entire spectral range, which is good indication for the absence of aggregation (Figure 8).

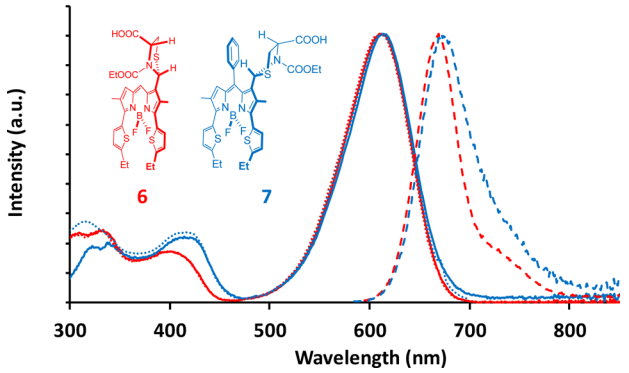

Figure 8. Absorption (plain trace), excitation (dotted trace), and emission (dashed trace) for compounds **6** (red) and **7** (blue) recorded in dichloromethane at 25 °C with c = 6.1 to 7.0 × 10^{-6} M for the absorption, c = 6.1 to 7.0 × 10^{-7} M for the excitation, and c = 6.1 to 7.0 × 10^{-7} M for the emission.

Optical Responses between 2b and 2c toward L-Cysteine. The time-dependent absorption and fluorescence responses of **2b** and **2c** in the presence of L-cysteine were investigated at 40 °C (Figure 9). The quantity of L-cysteine (100 equiv) added for the experiments was defined as the quantity required for the absorbance signal to reach the saturation value within 8 h. The absorption spectra were recorded every hour and the emission spectra every 30 min.

Changes in the absorption spectrum of BODIPY **2b** during reaction with L-cysteine in saline phosphate buffer (PBS) at pH = 7.4/EtOH (1:9, v/v) are shown in Figure 9a, and the same results for the fluorescence spectra are shown in Figure 9b. The absorption band centered at 631 nm was lost and became replaced by another at 605 nm, with an isosbestic point at 628 nm. In fluorescence, the emission band centered at 716 nm shifted to 669 nm, with an isoemissive point at 704 nm. Bands in the absorption and emission spectra of the thiazolidine compound **5** have to be of the same origin as those of the N-protected thiazolidine compound **6** (Figure 8). For compound **5**, λ_{abs} = 605 nm and λ_{em} = 669 nm compared nicely with 596 and 650 nm, respectively, for compound **6** in EtOH.

Thus, the changes seen in Figure 9 are consistent with the formation of the thiazolidine resulting from the cycloaddition of L-cysteine to the aldehyde units. In addition, the changes in absorption spectra of BODIPY **2c** on reaction with L-cysteine in DMSO are shown in Figure 9c, and those in the fluorescence spectra are shown in Figure 9d. Here, the initial absorption band centered at 633 nm shifted to one centered at 612 nm, with an isosbestic point at 631 nm. In the fluorescence spectra, a band at 676 nm grew at the expense of that at 718 nm, with an isoemissive point at 685 nm. Again λ_{abs} and λ_{em} of the cycloadduct were expected to be similar to the protected thiazolidine derivative **7**, which indeed is the case (with λ_{abs} 612 nm instead of 616 nm and for λ_{em} 676 nm instead of 673 nm).

In both cases, these ratiometric responses in the presence of L-cysteine were associated with a visually detectable change of the solution color from light blue to ultramarine blue (Figure 9a). Qualitatively, the changes are more obvious by fluorescence because the initial near-infrared emission at 716 nm is not detectable by eye, but the shift to 669 nm due to the reaction makes the red emission (Figure 9b) visible under irradiation from a simple bench UV lamp.

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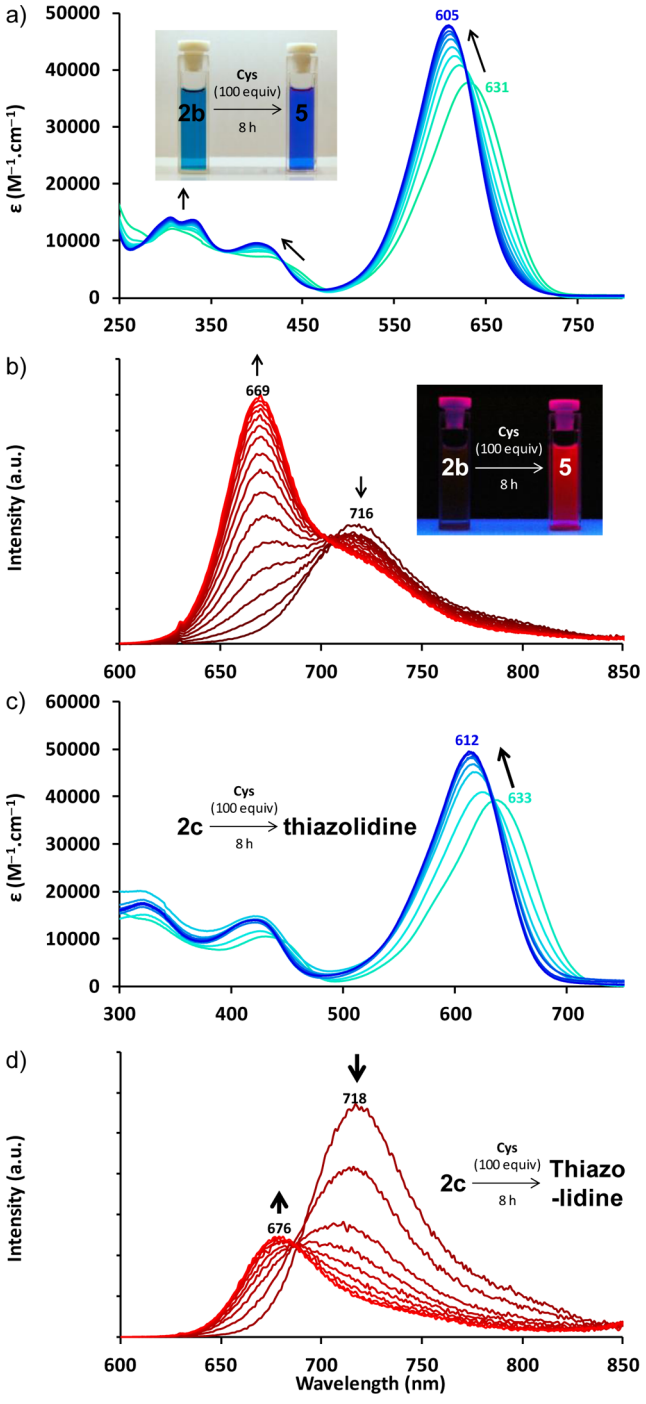

Figure 9. Time-dependent spectroscopy. (a) Evolution of the absorption properties of BODIPY **2b** ($c = 1.1 \times 10^{-5}$) in the presence of 100 equiv of L-Cys in pH 7.4 PBS/EtOH (1:9, v/v) at 40 °C upon 8 h (inset: visualized image of **2b** at the beginning and after 8 h under visible light). (b) Evolution of the fluorescence properties of **2b** under the same conditions described in part a. Excitation wavelength at 628 nm (inset: visualized image of **2b** at the beginning and after 8 h under UV lamp). Evolution of the (c) absorption and (d) emission properties of BODIPY **2c** ($c = 4.6 \times 10^{-5}$) in the presence of 100 equiv of L-Cys in DMSO at 40 °C after 8 h. Excitation wavelength at 631 nm.

CONCLUSIONS

By judicious substitution of the thienyl rings in dithienyl-BODIPY derivatives, formylation can be achieved in a largely regiospecific manner. The Vilsmeier–Haack reaction occurs regioselectively on the thiophene ring if the position α to the sulfur is free. If this position is blocked by an ethyl group, the reaction takes place exclusively at the β'-position of the BODIPY platform. With a phenyl group in the meso-position, a mixture of the β'-formylated compound (minor compound) and a 3-thienylaldehyde, **4c**, was produced. Two of these β'-formylated dyes, **2b** and **2c**, reacted with L-cysteine to give thiazolidine cycloadducts, which after N-protection provided thiazolidines **6** and **7** which could be separated, purified, and thus fully characterized. The NMR spectra of these compounds show that N-protection via carbamate formation involves C(2) inversion and leads selectively to the formation of the cis diastereoisomer.

The shift in the emission wavelength of the BODIPY-aldehyde chromophores **2b** and **2c** resulting from the formation of thiazolidine derivatives allows their emission to become readily observed by the naked eye. Thus, **2b** and **2c** appear to be interesting candidates for the detection of cysteine under physiological conditions. The ratiometric detection process in the therapeutic window could have interesting applications for sensing purposes. The kinetics of formation of the thiazolidine are slow, but by chemical tailoring of the BODIPY core it is expected that the interaction with mercapto biomolecules and thiols under biological conditions could be enhanced by adding supramolecular interactions, for example, between the carboxyl group of the L-cysteine and the reactive fluorescent platform. Work along these lines is currently in progress.

EXPERIMENTAL SECTION

General Methods. ^{1}H and ^{13}C spectra were recorded at rt on 300 and 400 MHz spectrometers using perdeuterated solvents as internal standards. Chemical shifts of ^{1}H and ^{13}C spectra are given in ppm relative to residual protiated solvent and relative to the solvent, respectively. ^{11}B spectra were recorded at rt on a 400 MHz spectrometer using $BF_3{\cdot}Et_2O$ as reference. FT-IR spectra were recorded using a spectrometer equipped with an ATR "diamond" apparatus. Chromatographic purification was conducted using 40–63 μm silica gel or aluminum oxide 90 standardized. Thin layer chromatography (TLC) was performed on silica gel or aluminum oxide plates coated with fluorescent indicator. All mixtures of solvents are given in v/v ratio. All anhydrous reactions were carried out under dry argon by using Schlenk tube techniques.

Spectroscopic Measurements. UV–visible spectra were recorded using a dual-beam grating spectrophotometer with a 1 cm quartz cell. All fluorescence spectra were corrected. The fluorescence quantum yield (Φ_{cmp}) was calculated from eq 1:

$$\Phi_{cmp} = \Phi_{ref}\frac{I}{I_{ref}}\frac{OD_{ref}}{OD}\frac{n^2}{n_{ref}^2} \quad (1)$$

Here, I denotes the integral of the corrected emission spectrum, OD is the optical density at the excitation wavelength, and n is the refractive index of the medium. Tetramethoxy-BODIPY, $\Phi_F = 0.49$ in dichloromethane ($\lambda_{exc} = 650$ nm), was used as reference.[33]

Luminescence lifetimes were measured on a spectrofluorimeter using software with time-correlated single photon mode coupled to a stroboscopic system. The excitation source was a laser diode ($\lambda = 310$ nm). No filter was used for the excitation. The instrument response function was determined by using a light-scattering solution (LUDOX).

Materials. All chemicals were used as received from commercial sources without further purification. CH_2Cl_2 was distilled over P_2O_5, and THF was distilled over sodium and benzophenone under an argon atmosphere. 3-Methyl-2-(thien-2-yl)-1*H*-pyrrole,[30a] **1a**,[30a] and **1b**[36] were synthesized according to the literature procedures.

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*4,4′-Difluoro-3,5-bis(5-ethylthienyl)-2,6-dimethyl-8-phenyl-4-bora-3a,4a-diaza-s-indacene (**1c**).* To a solution of 3-methyl-2-(thien-2-yl)-1*H*-pyrrole (1.61 g, 8.4 mmol, 2 equiv) in dry CH_2Cl_2 (60 mL) were successively added triethyl orthobenzoate (0.95 mL, 4.2 mmol, 1 equiv) and trifluoroacetic acid (0.32 mL, 4.2 mmol, 1 equiv). The mixture was stirred at rt overnight. Then triethylamine (3.5 mL, 25.2 mmol, 6 equiv) was added. After 10 min of stirring, $BF_3{\cdot}Et_2O$ (4.2 mL, 33.7 mmol, 8 equiv) was added. The solution was stirred at rt for 3 h, poured into a saturated solution of $NaHCO_3$, and stirred further for 1 h (2 × 100 mL). The organic layer was washed with water and with brine and dried over $MgSO_4$. The residue was purified by column chromatography (SiO_2, petroleum ether/CH_2Cl_2 60:40). Compound **1c** was recrystallized from CH_2Cl_2/EtOH and obtained as gold metallic crystals in 45% yield (979 mg): mp 251 °C; ^{1}H NMR (300 MHz, $CDCl_3$, ppm): δ = 7.64 (d, ^3J = 3.7 Hz, 2H), 7.52 (m, 5H), 6.86 (d, ^3J = 3.7 Hz, 2H), 6.59 (s, 2H), 2.91 (q, ^3J = 7.5 Hz, 4H), 2.19 (s, 6H), 1.36 (t, ^3J = 7.5 Hz, 6H); ^{13}C NMR (75 MHz, $CDCl_3$, ppm): δ = 151.3, 149.5, 139.4, 134.8, 134.5, 131.9 (t, J_{C-F} = 5.8 Hz), 130.5, 129.9, 129.6, 128.1, 124.3, 23.5, 15.5, 13.6; ^{11}B NMR (128 MHz, $CDCl_3$, ppm): δ = 1.23 (t, ^1J = 31.9 Hz); UV−vis (CH_2Cl_2) λ nm (ε, M^{-1} cm^{-1}): 616 (54000), 417 (14000), 324 (22000); IR (ν, cm^{-1}): 2967, 2925, 2873, 1576, 1553, 1471, 1405; EI-MS *m*/*z* (nature of the peak, relative intensity): 516.2 ([M]$^+$, 100). Anal. Calcd for $C_{29}H_{27}BF_2N_2S_2$: C, 67.44; H, 5.27; N, 5.42. Found: C, 67.18; H, 5.01; N, 5.11.

*1-Formyl-4,4′-difluoro-2,6-dimethyl-3,5-di(thien-2-yl)-4-bora-3a,4a-diaza-s-indacene (**2a**) and 4,4′-Difluoro-3-(5-formylthien-2-yl)-2,6-dimethyl-5-(thien-2-yl)-4-bora-3a,4a-diaza-s-indacene (**3a**).* Under argon, a solution of $POCl_3$ (3.2 mL) in DMF (3.2 mL) was stirred at 0 °C for 5 min and then at rt for 30 min. **1a** (100 mg, 0.26 mmol, 1 equiv) in $C_2H_4Cl_2$ (40 mL) was added, and the reaction mixture was stirred at 50 °C overnight. The reaction mixture was cooled and poured into a cold saturated solution of $NaHCO_3$ (140 mL). The resulting mixture was stirred for 1 h further, and the organic layer was washed with water and dried over $MgSO_4$. The solvent was removed under vacuum. The residue was purified by column chromatography (SiO_2, petroleum ether/CH_2Cl_2 40:60).

Compound **2a** was recrystallized from CH_2Cl_2/EtOH and obtained as green metallic needles in 9% yield (10 mg): mp 210 °C; ^{1}H NMR (300 MHz, $CDCl_3$, ppm): δ = 10.23 (s, 1H), 8.01 (dd, ^3J = 4.0 Hz, ^4J = 1.2 Hz, 1H), 7.81 (s, 1H), 7.64 (dd, ^3J = 5.0 Hz, ^4J = 1.0 Hz, 1H), 7.51 (dd, ^3J = 5.1 Hz, ^4J = 1.2 Hz, 1H), 7.48 (d, ^3J = 3.6 Hz, 1H), 7.19 (dd, ^3J = 4.0 Hz, ^3J = 5.0 Hz, 1H), 7.15 (dd, ^3J = 3.6 Hz, ^3J = 5.1 Hz, 1H), 7.05 (s, 1H), 2.38 (s, 3H), 2.31 (s, 1H); ^{13}C NMR (75 MHz, $CDCl_3$, ppm): δ = 186.7, 156.6, 144.3, 138.7, 134.3 (t, J_{C-H} = 7.1 Hz), 134.12, 134.09, 133.4, 131.84, 131.76, 131.3, 131.2, 131.1 (t, J_{C-H} = 4.4 Hz), 129.5, 128.4, 128.3, 127.3, 124.7, 14.3, 10.2; ^{11}B NMR (128 MHz, $CDCl_3$, ppm): δ = 1.13 (t, ^1J = 31.8 Hz); UV−vis (CH_2Cl_2) λ nm (ε, M^{-1} cm^{-1}): 624 (49000), 396 (10000), 306 (12000); IR (ν, cm^{-1}): 3118, 3071, 2962, 2915, 2859, 1664, 1611, 1514, 1426, 1396; EI-MS *m*/*z* (nature of the peak, relative intensity): 412.0 ([M]$^+$, 100), 393.1 ([M − F]$^+$, 15). Anal. Calcd for $C_{20}H_{15}BF_2N_2OS_2$: C, 58.26; H, 3.67; N, 6.79. Found: C, 58.12; H, 3.51; N, 6.57.

Compound **3a** was recrystallized from CH_2Cl_2/EtOH and obtained as green metallic needles in 65% yield (70 mg): mp 194 °C; ^{1}H NMR (300 MHz, $CDCl_3$, ppm): δ = 9.93 (s, 1H), 7.86 (dd, ^3J = 3.8 Hz, ^4J = 1.2 Hz, 1H), 7.82 (d, ^3J = 4.1 Hz, 1H), 7.76 (d, ^3J = 4.1 Hz, 1H), 7.58 (dd, ^3J = 5.1 Hz, ^4J = 1.2 Hz, 1H), 7.19 (dd, ^3J = 3.8 Hz, ^3J = 5.1 Hz, 1H), 7.05 (s, 1H), 6.93 (s, 1H), 6.83 (s, 1H), 2.25 (s, 3H), 2.24 (s, 3H); ^{13}C NMR (75 MHz, $CDCl_3$, ppm): δ = 183.0, 153.1, 145.6, 144.5, 142.0, 136.3, 136.1, 134.7, 132.7, 132.01, 132.00, 131.8, 131.7, 131.2, 130.0, 128.5, 127.9, 125.5, 13.8, 13.3; ^{11}B NMR (128 MHz, $CDCl_3$, ppm): δ = 1.22 (t, ^1J = 31.8 Hz); UV−vis (CH_2Cl_2) λ nm (ε, M^{-1} cm^{-1}): 604 (51000), 359 (17000), 337 (17000); IR (ν, cm^{-1}): 3101, 3089, 2947, 2916, 2796, 1669, 1604, 1522, 1488, 1445, 1411, 1395, 1376; EI-MS *m*/*z* (nature of the peak, relative intensity): 412.0 ([M]$^+$, 100); Anal. Calcd for $C_{20}H_{15}BF_2N_2OS_2$: C, 58.26; H, 3.67; N, 6.79. Found: C, 58.04; H, 3.38; N, 6.51.

*3,5-Bis(5-ethylthien-2-yl)-1-formyl-4,4′-difluoro-2,6-dimethyl-4-bora-3a,4a-diaza-s-indacene (**2b**).* Under argon, a solution of $POCl_3$ (11 mL) in DMF (11 mL) was stirred at 0 °C for 5 min and then at rt for 30 min. **1b** (400 mg, 0.91 mmol, 1 equiv) in $C_2H_4Cl_2$ (80 mL) was added, and the reaction mixture was stirred at 50 °C overnight. The reaction mixture was cooled and poured into a cold saturated solution of $NaHCO_3$ (70 mL). The resulting mixture was stirred for 1 h further, and the organic layer was washed with water and dried over $MgSO_4$. The solvent was removed under vacuum. The residue was purified by column chromatography (SiO_2, petroleum ether/CH_2Cl_2 40:60). Compound **2b** was recrystallized from CH_2Cl_2/EtOH and obtained as blue metallic crystals in 85% yield (362 mg): mp 223 °C; ^{1}H NMR (300 MHz, $CDCl_3$, ppm): δ = 10.19 (s, 1H), 7.97 (d, ^3J = 4.0 Hz, 1H), 7.69 (s, 1H), 7.33 (d, ^3J = 3.6 Hz, 1H), 6.98 (s, 1H), 6.91 (d, ^3J = 4.0 Hz, 1H), 6.84 (d, ^3J = 3.6 Hz, 1H), 2.92 (q, ^3J = 7.4 Hz, 2H), 2.91 (q, ^3J = 7.4 Hz, 2H), 2.39 (s, 3H), 2.30 (s, 3H), 1.36 (t, ^3J = 7.4 Hz, 3H), 1.35 (t, ^3J = 7.4 Hz, 3H); ^{13}C NMR (75 MHz, $CDCl_3$, ppm): δ = 186.6, 156.4, 155.6, 150.5, 144.1, 138.7, 135.2 (t, J_{C-H} = 7.7 Hz), 134.0, 133.4, 133.3, 131.2, 131.0 (t, J_{C-H} = 3.6 Hz), 129.0, 128.9, 128.7, 125.6, 123.8, 123.0, 23.6, 23.4, 15.43, 15.37, 14.7, 10.4; ^{11}B NMR (128 MHz, $CDCl_3$, ppm): δ = 1.23 (t, ^1J = 31.8 Hz); UV−vis (CH_2Cl_2) λ nm (ε, M^{-1} cm^{-1}): 643 (50000), 415 (9000), 312 (13000); IR (ν, cm^{-1}): 3075, 2965, 2928, 2740, 1667, 1609, 1562, 1530, 1488, 1463, 1452, 1427, 1395; EI-MS *m*/*z* (nature of the peak, relative intensity): 468.1 ([M]$^+$, 100), 449.1 ([M − F]$^+$, 100). Anal. Calcd for $C_{24}H_{23}BF_2N_2OS_2$: C, 61.54; H, 4.95; N, 5.98. Found: C, 61.37; H, 4.61; N, 5.61.

*4,4′-Difluoro-1-formyl-2,6-dimethyl-3,5-bis(5-ethylthien-2-yl)-8-phenyl-4-bora-3a,4a-diaza-s-indacene (**2c**) and 4,4′-Difluoro-2,6-dimethyl-3-[5-ethyl-4-formylthien-2-yl]-5-(5-ethylthien-2-yl)-8-phenyl-4-bora-3a,4a-diaza-s-indacene (**4c**).* Under argon, a solution of $POCl_3$ (2.4 mL) in DMF (2.4 mL) was stirred at 0 °C for 5 min and then at rt for 30 min. **1c** (100 mg, 0.19 mmol, 1 equiv) in $C_2H_4Cl_2$ (10 mL) was added, and the reaction mixture was stirred at 50 °C overnight. The reaction mixture was cooled and poured into a cold saturated solution of $NaHCO_3$ (100 mL). The resulting mixture was stirred for 1 h further, and the organic layer was washed with water and dried over $MgSO_4$. The solvent was removed under vacuum. The residue was purified by column chromatography (SiO_2, petroleum ether/CH_2Cl_2 50:50).

Compound **2c** was recrystallized from CH_2Cl_2/EtOH and obtained as red metallic needles in 22% yield (23 mg): mp 199 °C; ^{1}H NMR (400 MHz, $CDCl_3$, ppm): δ = 8.71 (s, 1H), 7.93 (d, ^3J = 3.9 Hz, 1H), 7.54 (m, 5H), 7.28 (d, ^3J = 3.5 Hz, 1H), 6.91 (d, ^3J = 3.9 Hz, 1H), 6.85 (d, ^3J = 3.5 Hz, 1H), 6.59 (s, 1H), 2.91 (q, ^3J = 7.5 Hz, 4H), 2.29 (s, 3H), 2.24 (s, 3H), 1.37 (t, ^3J = 7.5 Hz, 3H), 1.36 (t, ^3J = 7.5 Hz, 3H); ^{13}C NMR (100 MHz, $CDCl_3$, ppm): δ = 188.0, 155.4, 154.9, 150.6, 145.4, 138.2, 137.7, 135.2, 134.8 (t, J_{C-F} = 7.3 Hz), 134.6, 133.1, 132.9, 132.3, 131.6, 131.2, 130.2, 130.0, 129.0, 128.7, 128.6, 125.5, 123.6, 23.6, 23.5, 15.43, 15.39, 14.5, 12.3; ^{11}B NMR (128 MHz, $CDCl_3$, ppm): δ = 1.06 (t, ^1J = 31.1 Hz); UV−vis (CH_2Cl_2) λ nm (ε, M^{-1} cm^{-1}): 636 (51000), 427 (13000), 323 (15000); IR (ν, cm^{-1}): 2963, 2925, 2873, 1659, 1546, 1496, 1451, 1322; EI-MS *m*/*z* (nature of the peak, relative intensity): 544.1 ([M]$^+$, 100). Anal. Calcd for $C_{30}H_{27}BF_2N_2OS_2$: C, 66.18; H, 5.00; N, 5.14. Found: C, 65.83; H, 4.72; N, 4.98.

Compound **4c** was recrystallized from CH_2Cl_2/EtOH and obtained as green metallic crystals in 45% yield (47 mg): mp 184 °C; ^{1}H NMR (400 MHz, $CDCl_3$, ppm): δ = 9.57 (s, 1H), 7.76 (d, ^3J = 3.9 Hz, 1H), 7.55 (m, 5H), 7.26 (s, 1H), 6.86 (d, ^3J = 3.9 Hz, 1H), 6.73 (s, 1H), 6.57 (s, 1H), 2.88 (q, ^3J = 7.5 Hz, 4H), 2.22 (s, 3H), 1.93 (s, 3H), 1.37 (t, ^3J = 7.5 Hz, 3H), 1.34 (t, ^3J = 7.5 Hz, 3H); ^{13}C NMR (100 MHz, $CDCl_3$, ppm): δ = 185.8, 153.7, 153.3, 149.8, 141.7, 141.6, 140.92, 140.86, 135.8, 134.4, 134.3, 133.6 (t, J_{C-F} = 7.0 Hz), 132.3, 131.9, 130.4, 130.2, 130.0, 129.0, 128.3, 126.4, 125.0, 120.91, 23.53, 23.30, 15.39, 14.93, 14.12, 11.47; ^{11}B NMR (128 MHz, $CDCl_3$, ppm): δ = 0.94 (t, ^1J = 31.5 Hz); UV−vis (CH_2Cl_2) λ nm (ε, M^{-1} cm^{-1}): 595 (53000), 394 (14000), 314 (15000); IR (ν, cm^{-1}): 2963, 2930, 2869, 2850, 1678, 1543, 1476, 1402, 1371; EI-MS *m*/*z* (nature of the peak, relative intensity): 544.1 ([M]$^+$, 100). Anal. Calcd for $C_{30}H_{27}BF_2N_2OS_2$: C, 66.18; H, 5.00; N, 5.14. Found: C, 66.04; H, 4.85; N, 5.17.

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*4,4′-Difluoro-1-[(4R)-4-carboxythiazolidin-2-yl]-2,6-dimethyl-3,5-bis(5-ethylthien-2-yl)-4-bora-3a,4a-diaza-s-indacene (**5**).* To a solution of **2b** (30 mg, 0.06 mmol, 1 equiv) in EtOH/PBS pH 7.4 (390 mL, 9:1) was added L-cysteine (78 mg, 0.64 mmol, 10 equiv). The solution was stirred at 40 °C for 20 h. The solvent was removed under vacuum. The residue was dissolved in AcOEt (30 mL) and washed with water. The organic layer was collected and dried over $MgSO_4$. The solvent was removed under vacuum. The residue was purified by crystallization from THF/pentane. Compound **5** was obtained as a blue dark powder in 55% yield (20 mg): ^{1}H NMR (300 MHz, DMSO-d_6, ppm): (mixture of two diastereoisomers) δ = 8.03 (s, 0.5H), 7.95 (s, 0.5H), 7.60 (d, ^4J = 3.8 Hz, 0.5H), 7.57 (d, ^4J = 3.8 Hz, 0.5H), 7.33 (t, ^4J = 4.6 Hz, 1H), 7.25 (s, 0.5H), 7.22 (s, 0.5H), 7.00 (t, ^4J = 4.0 Hz, 1H), 6.96 (m, 1H), 5.92 (s, 0.5H), 5.85 (s, 0.5H), 4.42 (m, 0.5H), 3.93 (m, 0.5H), 3.32 (m, 2H), 2.88 (q, ^3J = 7.4 Hz, 4H), 2.22 (s, 1.5H), 2.21 (s, 1.5H), 2.17 (s, 1.5H), 2.10 (s, 1.5H), 1.29 (t, ^3J = 7.4 Hz, 3H), 1.28 (t, ^3J = 7.4 Hz, 3H).

*4,4′-Difluoro-1-[(4R)-4-carboxy-3-(ethoxycarbonyl)thiazolidin-2-yl]-2,6-dimethyl-3,5-bis(5-ethylthien-2-yl)-4-bora-3a,4a-diaza-s-indacene (**6**).* To a solution of **2b** (30 mg, 0.06 mmol, 1 equiv) in EtOH/PBS pH 7.4 (390 mL, 9:1) was added L-cysteine (116 mg, 0.96 mmol, 15 equiv). The solution was stirred at 40 °C for 20 h. The solvent was removed under vacuum. The residue was dissolved in AcOEt (30 mL) and washed with water. The organic layer was collected and dried over $MgSO_4$. The solvent was removed under vacuum. The residue was dissolved in dry THF (10 mL). To this mixture was added dropwise ethyl chloroformate (37 μL, 0.38 mmol, 6 equiv). The solution was stirred at rt for 1 h, and the solvent was removed under vacuum. The residue was purified by column chromatography (SiO_2, CH_2Cl_2 to CH_2Cl_2/EtOH 100:0 to 70:30, 0.1% of AcOH. Compound **6** was obtained as a blue powder in 63% (25 mg): ^{1}H NMR (300 MHz, $CDCl_3$, ppm): δ = 8.34 (s, 1H), 7.57 (d, ^4J = 3.6 Hz, 1H), 7.28 (d, ^4J = 3.6 Hz, 1H), 6.85 (s, 1H), 6.75 (t, ^4J = 4.1 Hz, 2H), 6.02 (s, 1H), 4.78 (t, ^3J = 4.5 Hz, 1H), 3.97 (m, 2H), 3.39 (m, 2H), 2.80 (q, ^3J = 7.5 Hz, 4H), 2.15 (s, 3H), 2.08 (s, 3H), 1.26 (t, ^3J = 7.5 Hz, 3H), 1.25 (t, ^3J = 7.5 Hz, 3H), 1.02 (m, 3H); ^{13}C NMR (75 MHz, $CDCl_3$, ppm): δ = 174.0, 172.2, 155.3, 151.8, 150.6, 147.1, 135.8, 135.0, 132.1 (t, J_{C-F} = 5.7 Hz), 131.6, 131.1, 130.6, 130.1, 129.6, 129.1, 127.2, 125.1, 124.4, 123.7, 62.4, 57.6, 33.2, 23.3, 20.5, 17.7, 15.2, 13.9, 13.6, 10.6; ^{11}B NMR (128 MHz, $CDCl_3$, ppm): δ = 1.15 (t, ^1J = 31.9 Hz); UV−vis (CH_2Cl_2) λ nm (ε, M^{-1} cm^{-1}): 610 (45000), 403 (9000), 334 (12000); IR (ν, cm^{-1}): 3105, 2966, 2922, 1708, 1596, 1466, 1401, 1376, 1305; EI-MS m/z (nature of the peak, relative intensity): 643.1 ([M]$^+$, 100). Anal. Calcd for $C_{30}H_{32}BF_2N_3O_4S_3$: C, 55.99; H, 5.01; N, 6.53. Found: C, 55.64; H, 4.89; N, 6.32.

*3,5-Bis(5-ethylthien-2-yl)-4,4′-difluoro-2,6-dimethyl-8-phenyl-1-[(4R)-4-carboxy-3-(ethoxycarbonyl)thiazolidin-2-yl]-4-bora-3a,4a-diaza-s-indacene (**7**).* To a solution of **2c** (19 mg, 0.04 mmol, 1 equiv) in THF/PBS pH 7.4 (20 mL, 9:1) was added L-cysteine (63 mg, 0.52 mmol, 15 equiv). The solution was stirred at 40 °C for 20 h. The solvent was removed under vacuum. The residue was dissolved in AcOEt (20 mL) and washed with water. The organic layer was collected and dried over $MgSO_4$. The solvent was removed under vacuum. The residue was dissolved in dry THF (10 mL). To this mixture was added dropwise ethyl chloroformate (20 μL, 0.21 mmol, 6 equiv). The solution was stirred at rt for 1 h, and the solvent was removed under vacuum. The residue was purified by column chromatography (SiO_2, CH_2Cl_2/EtOH 100:0 to 90:10, 0.1% of AcOH). Compound **7** was obtained as a blue powder in 40% yield (10 mg): ^{1}H NMR (400 MHz, $CDCl_3$, ppm): δ = 7.62 (d, ^3J = 3.8 Hz, 1H), 7.41 (m, 5H), 7.21 (d, ^3J = 3.7 Hz, 1H), 6.78 (d, ^3J = 3.8 Hz, 1H), 6.76 (d, ^3J = 3.7 Hz, 1H), 6.24 (s, 1H), 4.77 (s, 1H), 4.44 (m, 1H), 3.98 (m, 2H), 3.28 (m, 1H), 2.83 (m, 4H), 2.64 (dd, ^3J = 7.1 Hz, ^3J = 7.1 Hz, 1H), 2.12 (s, 3H), 2.09 (s, 3H), 1.28 (t, ^3J = 7.4 Hz, 3H), 1.27 (t, ^3J = 7.4 Hz, 3H), 1.00 (t, ^3J = 7.1 Hz, 3H); ^{13}C NMR (100 MHz, $CDCl_3$, ppm): δ = 170.9, 159.8, 152.1, 150.7, 150.6, 149.6, 138.8, 136.3, 135.6, 134.5, 132.4 (t, J_{C-F} = 7.0 Hz), 131.3, 131.2, 130.8, 130.3, 130.2, 129.4, 129.2, 128.9, 128.7, 128.2, 127.9, 124.5, 123.5, 63.8, 62.5, 60.3, 32.0, 23.34, 23.28, 15.3, 14.0, 13.6, 11.2; ^{11}B NMR (128 MHz, $CDCl_3$, ppm): δ = 0.95 (t, ^1J = 31.5 Hz); UV−vis (CH_2Cl_2) λ nm (ε, M^{-1} cm^{-1}): 615 (48000), 415 (12000), 340 (11000); EI-MS m/z (nature of the peak, relative intensity): 719.1 ([M]$^+$, 100). Anal. Calcd for $C_{36}H_{36}BF_2N_3O_4S_3$: C, 60.08; H, 5.04; N, 5.84. Found: C, 59.82; H, 4.70, N, 5.72.

■ ASSOCIATED CONTENT

Supporting Information

Traces of the NMR spectra (proton, carbon, boron), COSY, and NOESY spectra for compounds **4c**, **6**, and **7** and spectroscopic data for compounds **1c**, **2a−c**, **3a**, **4c**, **6**, **7** in different solvents. This material is available free of charge via the Internet at http://pubs.acs.org.

■ AUTHOR INFORMATION

Corresponding Authors

*E-mail: denicola@unistra.fr.

*E-mail: ziessel@unistra.fr.

Notes

The authors declare no competing financial interest.

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