Thin-film ratiometric fluorescent chemosensors with tunable performance characteristics

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Supporting Information

Additional Figures.

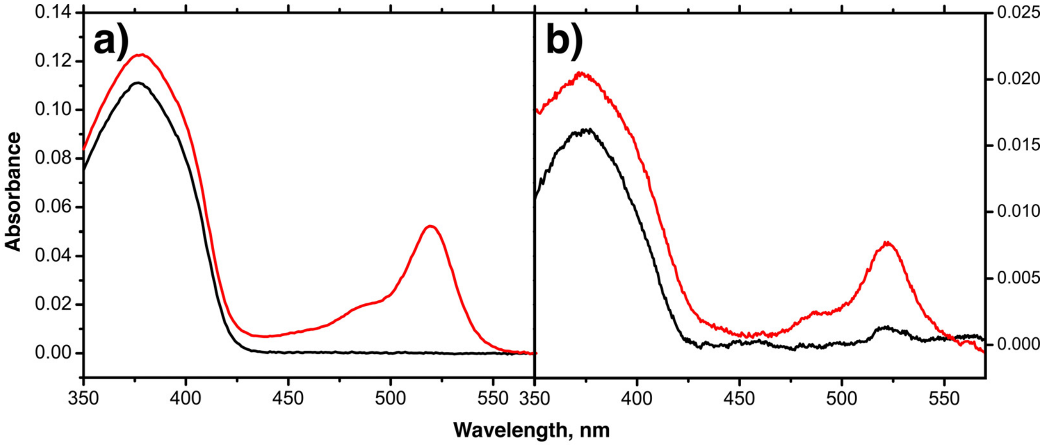


Figure S1. a) UV/vis absorption spectra of the 15 μM solution of **1** in THF before *(black trace)* and after *(red trace)* addition of 2 equivalents of Bu_4NF. b) UV/vis absorption spectra of the monolayer of **1** on glass before *(black trace)* and after *(red trace)* 3 min exposure to 9.0 μM solution of Bu_4NF in THF.

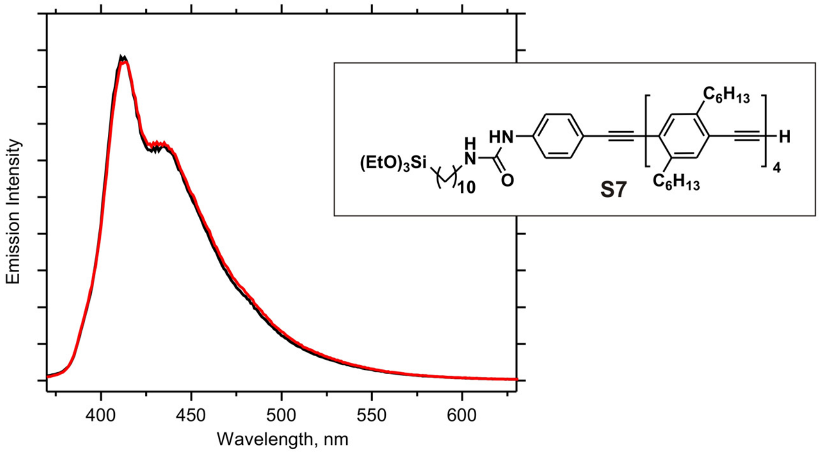


Figure S2. Emission spectra of the glass-immobilized monolayer of PE-only reference compound **S7**[1] before *(black trace)* and after *(red trace)* exposure to 9.0 μM solution of Bu_4NF in THF for 3 min. The absence of change in the spectra demonstrates integrity of the monolayer upon exposure to fluoride solution.

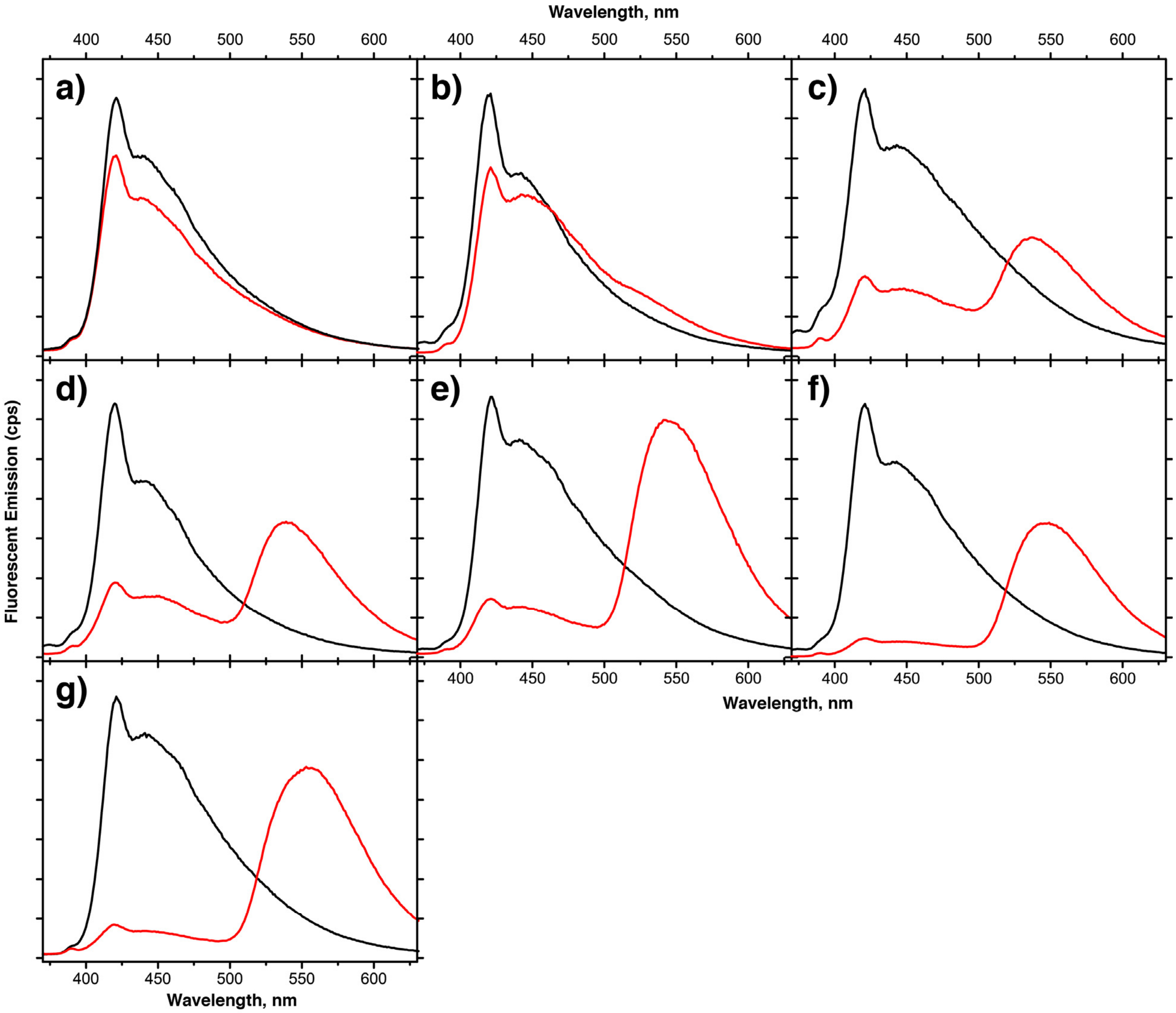


Figure S3. Fluorescence spectra of glass-immobilized monolayer of **1** prior to exposure to fluoride (*black traces*) and after exposure to Bu_4NF solutions in THF *(red traces)* with the following fluoride concentrations: 5.0 μM (a), 6.0 μM (b), 6.5 μM (c), and 7.0 μM (d), 7.5 μM (e), 9.0 μM (f), and 10.0 μM (g). Spectra were acquired at 350 nm irradiation.

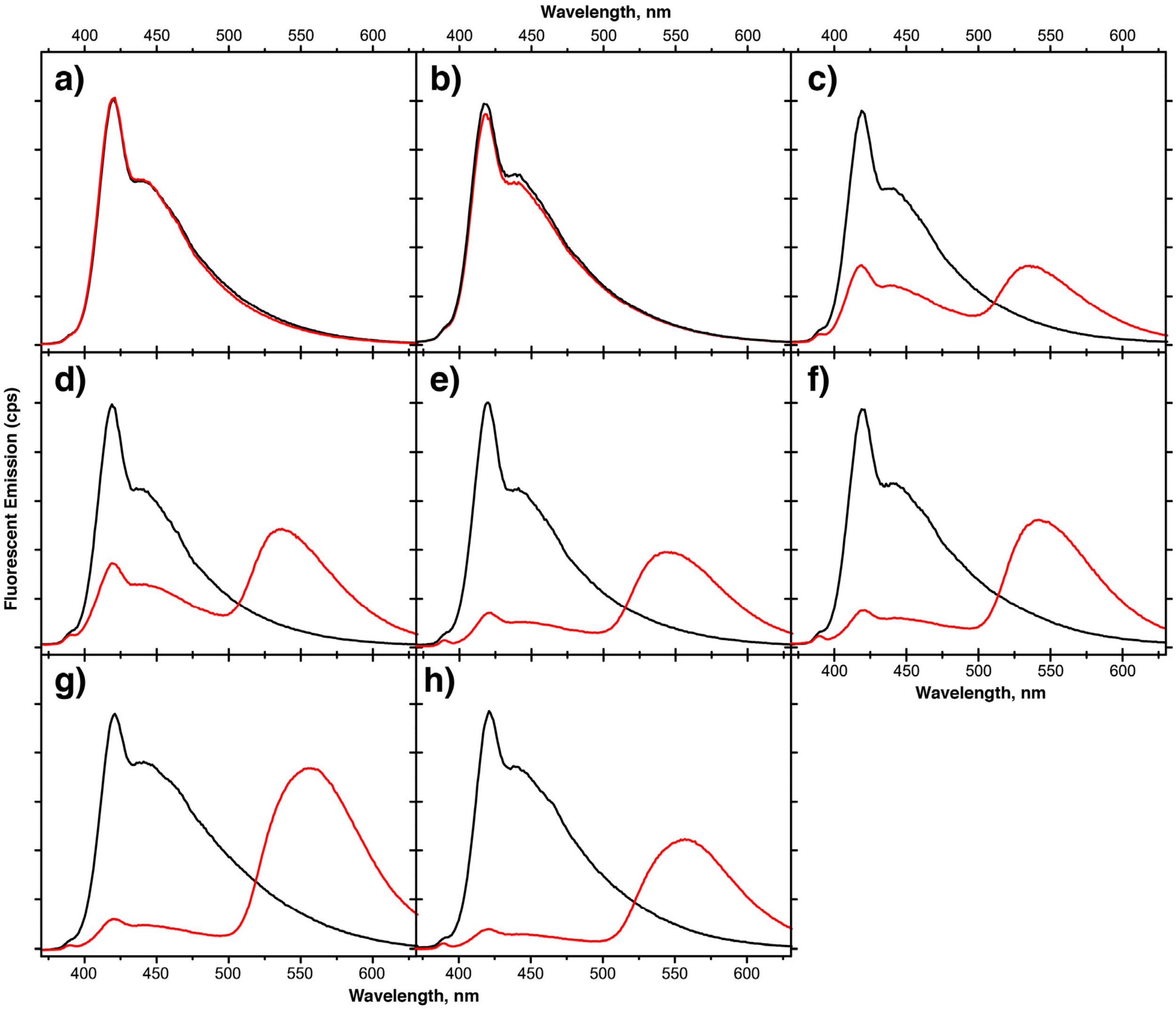


Figure S4. Fluorescence spectra of glass-immobilized monolayer of **1** prepared by co-deposition from solution of **1** and ODMS in 1:4 ratio prior to exposure to fluoride (*black traces*) and after exposure to Bu_4NF solutions in THF *(red traces)* with the following fluoride concentrations: 4.0 µM (a), 5.0 µM (b), 6.0 µM (c), and 7.0 µM (d), 9 µM (e), 10.0 µM (f), 12.0 µM (g), and 14.0 µM. Spectra were acquired at 350 nm irradiation.

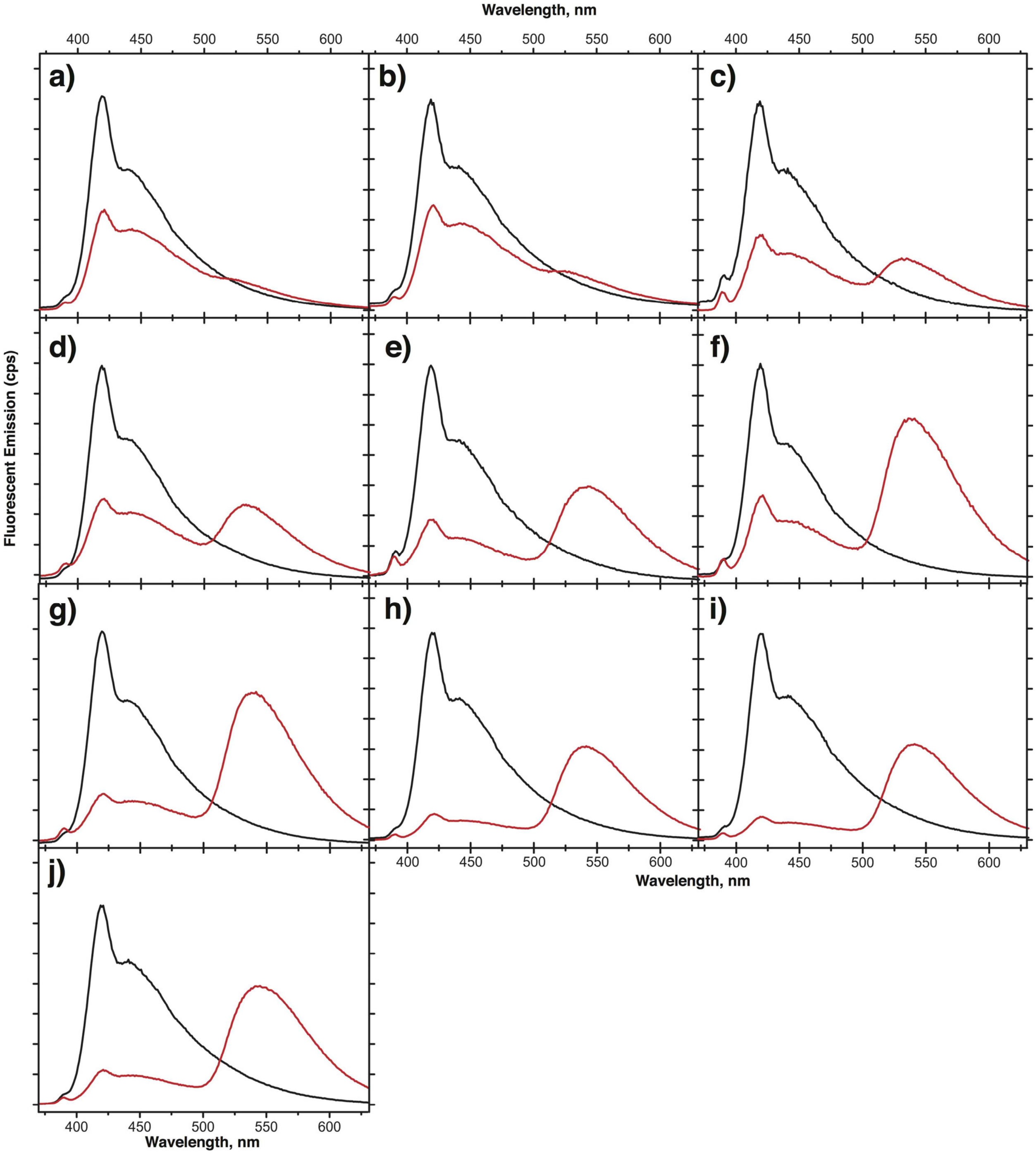


Figure S5. Fluorescence spectra of glass-immobilized monolayer of **1** prepared by co-deposition from solution of **1** and ODMS in 1:8 ratio prior to exposure to fluoride (*black traces*) and after exposure to Bu_4NF solutions in THF *(red traces)* with the following fluoride concentrations: 4.0 μM (a), 5.0 μM (b), 7 μM (c), 9 μM (d), 11.0 μM (e), 12 μM (f), 14 μM (g), 15 μM (h), 16.0 μM (i), and 17.0 μM (j). Spectra were acquired at 350 nm irradiation.

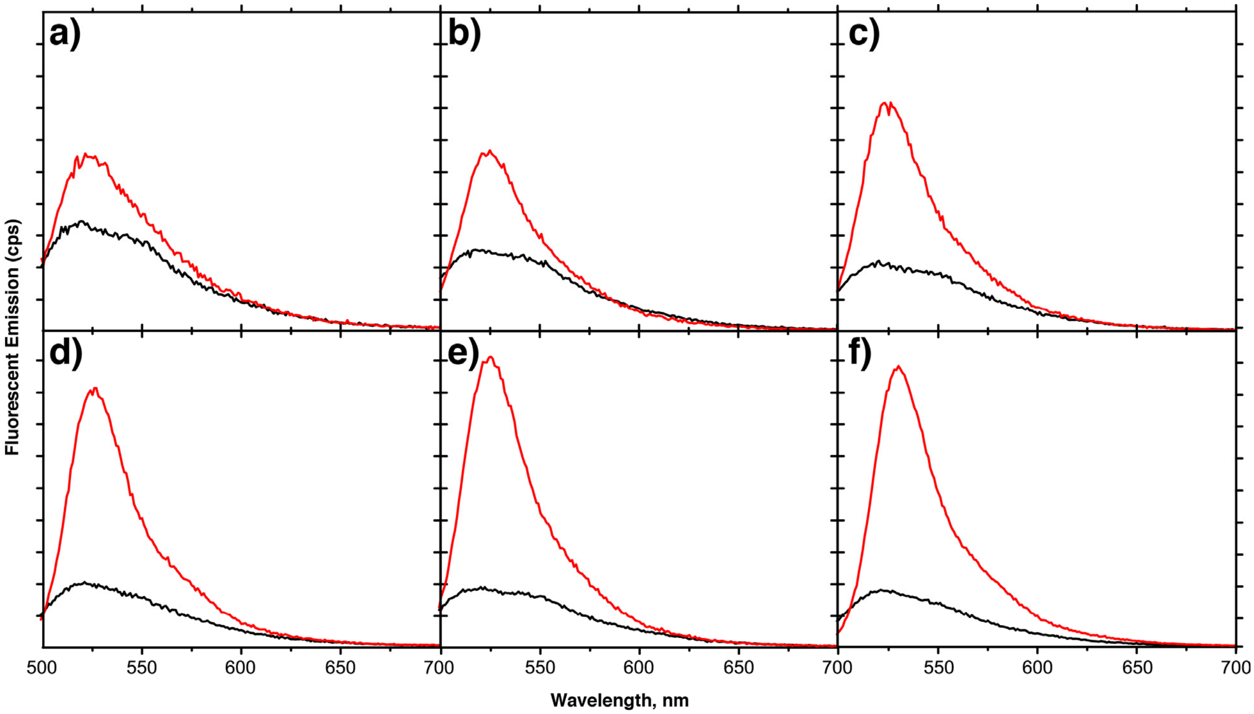


Figure S6. Fluorescence spectra of glass-immobilized monolayer of **2** prior to exposure to fluoride (*black traces*) and after exposure to Bu_4NF solutions in THF *(red traces)* with the following fluoride concentrations: 1.83 μM (a), 4.17 μM (b), 6.25 μM (c), 8.00 μM (d), 10.00 μM (e), and 12.0 μM (f). Spectra were acquired at 475 nm irradiation.

Experimental Details.

General Procedures. All reactions were performed under an atmosphere of dry nitrogen, except those that required Schlenk techniques, which were performed under an atmosphere of ultrapure argon. Chromatographic separations were carried out on silica gel (EMD, 60 Å, 40-63 μM, pH 6.8-7.0) slurry packed into glass columns. Toluene, hexane and THF were dried by passing through columns of activated alumina and *N,N*-dimethylformamide (DMF) was dried by passing through a column of molecular sieves using a PS-400 Solvent Purification System from Innovative Technologies, Inc. The water content in the solvents was confirmed by coulometric titration (using a DL32 coulometric titrator from Mettler Toledo). High quality $Pd(PPh_3)_4$ was obtained from Strem Chemicals, Inc., while all other reagents were obtained from Sigma-Aldrich and Alfa Aesar and used as received. Indium tin oxide (ITO) coated glass with an 8-12 Ω/sq. surface resistivity was purchased from Delta Technologies, Ltd. ^1H NMR spectra were recorded at 250 MHz and are reported in parts per million downfield from tetramethylsilane. UV-Visible spectra were recorded either on a Varian Cary 50 (solutions) or an Agilent Cary 5000 (monolayers) spectrophotometer. Fluorescence studies were carried out using a PTI QuantaMaster4/2006SE spectrofluorimeter. Quantum yields in monolayers were estimated relative to ~1 mM 9,10-diphenylanthracene in PMMA spun-cast film ($\Phi = 0.83^2$). High-resolution mass spectra were obtained at the Mass Spectrometry Facility at LSU Chemistry Department using an ESI-TOF or MALDI-TOF method with peak matching protocol to determine the mass and error range of the molecular ion.

Cleaning and Activation of Glass Slides. Microscope cover slips (25×25mm, Slip-Rite No. 1) were cut using a ceramic cutting tile to fit tightly across the diagonal of a 1 cm cuvette. The glass pieces were washed subsequently with acetone, chloroform, methanol, and then water for 30 minutes each under sonication. The slides were then thoroughly dried under a flow of nitrogen. The cleaned slides were placed in 20 ml scintillation vials which were filled with freshly prepared piranha solution (conc. H_2SO_4 and 30% H_2O_2 in 7:3 ratio). (***NOTE:*** *extreme care must be taken when dealing with piranha solutions as they can detonate when contacted with organic compounds!*) The vials were kept at 75 °C for 1 h, then were allowed to cool to room temperature, and the slide were removed and rinsed with copious amounts of Millipore-pure water. The rinsed slides were dried under a flow of nitrogen overnight and then used for immobilization. ITO/glass slides was activated using "basic piranha" (NH_4OH : H_2O_2 : Water, 1:1:5) and prewashed using the same procedure as the glass slides.

Monolayer Preparation Procedure. A 0.1 mM stock solution of **1** was prepared in dry toluene. For monolayer "dilution" studies, three stock solutions of octadecyl(triethoxy)methylsilane (ODMS) were prepared with concentrations 0.8 mM, 1.6 mM, and 2.4 mM. Scintillation vials were filled with 8 ml of the stock solution of **1** and 4 ml of each of the three ODMS stock solutions were added to end up with 1:4, 1:8, and 1:12 molar ratio of **1** to ODMS. For slides with un-dilute monolayer of **1**, 4 ml of toluene was added to 8 ml of the stock solution of **1**. Thus, all series of deposition solutions maintained a uniform sensor **1** concentration of 0.06 mM. Two freshly activated slides were carefully placed in each scintillation vial with deposition solutions in such a way that they did not fall on top of each other, and the vials were kept at 70 °C for 1 h. The slides were then removed and rinsed with copious toluene, and sonicated for 30 min in toluene, followed by a second copious toluene rinse and drying under nitrogen.

Fluoride Detection Procedure. Freshly prepared slides were immersed in 0.4 mM solution of Bu_4NPF_6 in THF for 30 seconds, and then placed in a spectrofluorimeter cuvette. The cuvette was filled with THF and the fluorescence spectrum of the unexposed film was acquired first. The solvent was carefully removed with a Pasteur pipette and replaced with a Bu_4NF solution of required concentration in THF. The fluoride solution was allowed to remain in the cuvette for 3 minutes, after which the emission spectrum of the fluoride-exposed film was recorded. Each slide was used to measure response to only one concentration of fluoride.

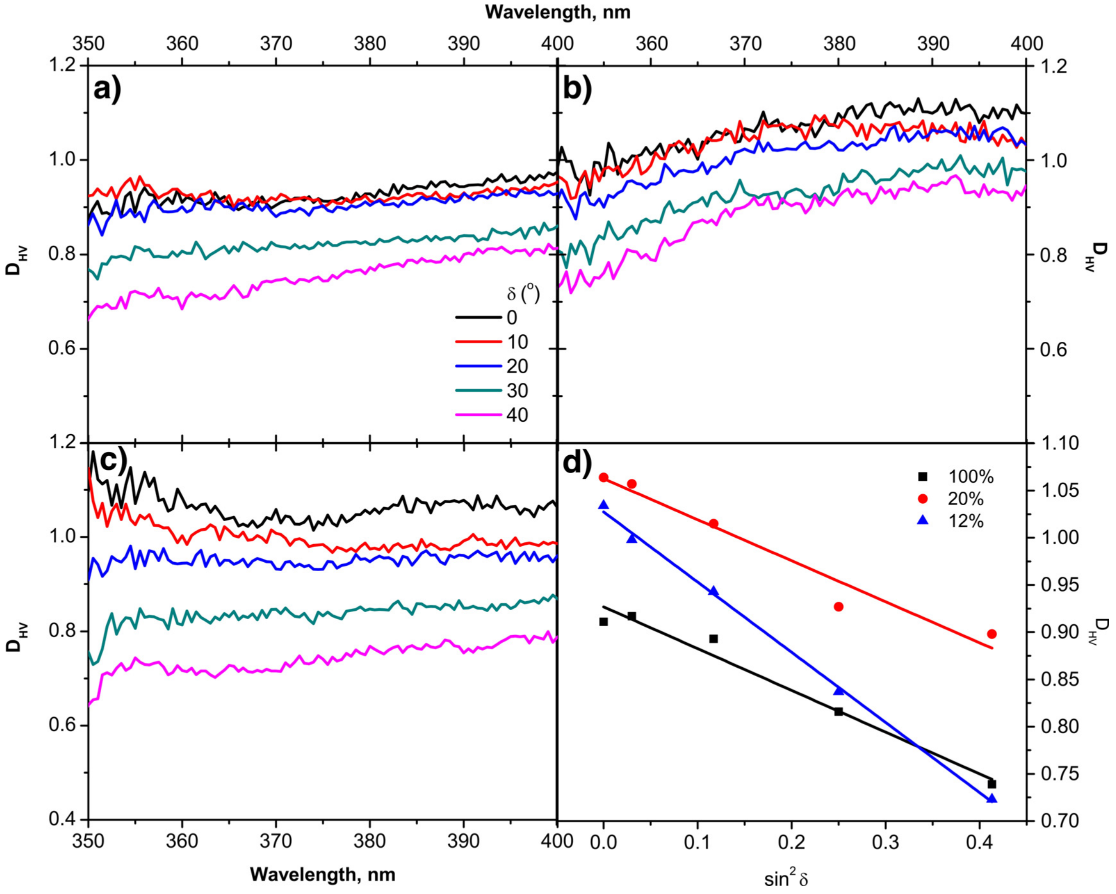


Figure S7. Wavelength dependence of the dichroic ratio D_{HV} at different twisting angles δ for "undiluted", 100% monolayer of **1** (a), and the monolayers prepared from solutions of **1** with ODMS "diluter" in molar ratio of 1:4 (b), and 1:8 (c). Linear relationship between D_{HV} at 370 nm and $\sin^2\delta$ for the monolayers in a-c (d).

Variable Angle Optical Anisotropy Measurements. To confirm the uniform alignment of the molecules of **1** in surface-immobilized monolayers, studies on optical anisotropy were carried out.[3] For these studies, UV/vis absorption spectra of the monolayers were obtained with vertically and horizontally polarized incident light, using an Agilent Cary 5000 spectrometer with a sample mounted on a Brewster angle holder, optical polarizer before the sample, and depolarizer after the sample. The twisting angle δ between the propagation direction of light and normal to the sample surface was changed in 10° increments from 0 to 40° (Figure S7a-c). The linear dependence between the dichroic ratio D_{HV} (ratio of sample absorbances with horizontally (A_{H}) and vertically (A_{V}) polarized light corrected by the spectrometer "G-factor") and $\sin^2\delta$ was required by the equation 1, and was indeed experimentally observed (Figure S7d). Furthermore, the linear dependence was not significantly affected by the monolayer dilution. This provided a strong evidence of unidirectional molecular alignment of the molecules of **1** in the monolayers.

$$D_{HV} = \frac{A_H}{A_V} = 1 + \frac{2 - 3\sin^2\varphi}{\sin^2\varphi}\sin^2\delta \qquad \text{(Eq. 1)}$$

In principle, from the slope of the plot in Figure S5d, and using the equation 1, one could estimate tilting angle φ between the surface normal and the direction of the transition dipole vector (which is expected to coincide with the long molecular axis) of the molecule of **1**. Nevertheless, such determinations are not highly reliable and thus we did not attempt to do this.

Evaluation of surface density of the monolayer of 1. Cyclic voltammetry measurements were performed using an Autolab PGSTAT 302 potentiostat. The measurements were carried out using a three-electrode system with monolayer-modified ITO/glass working electrode (electrode area ~1.13 cm^2), Ag/AgNO$_3$ non-aqueous reference electrode, and a Pt gauze counter electrode in 0.1 M Bu$_4$NPF$_6$ solution in CH$_2$Cl$_2$ as supporting electrolyte. The reference electrode was checked against ferrocene standard every time before and after the experiments were performed, and the measured potentials were corrected based on the Fc/Fc$^+$ redox potential value.

The surface coverage density was estimated based on the measured area of the redox peak corresponding to irreversible oxidation of fluorescein group at ~0.9 V (Figure S7). The value was corrected by the corresponding data for the bare ITO electrode. Assuming the reduction being a one-electron process, the surface density Γ was estimated using the formula:

$$\Gamma = \frac{Q}{F}$$

where Q is the redox peak area (C cm^{-2}), and F is Faraday constant (96500 C mol^{-1}). For the current case ($Q = 4.98\times10^{-6}$ C cm^{-2} after correcting for bare ITO electrode), the surface density Γ was estimated as 5.17×10^{-11} mol cm^{-2}, which translates to 3.2 nm^2 per molecule of **1**.

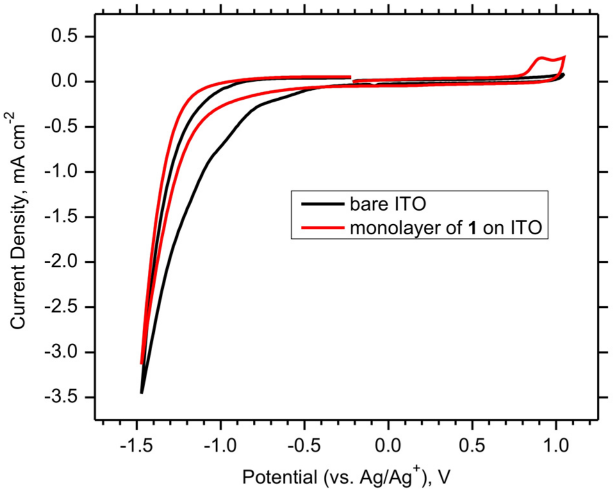
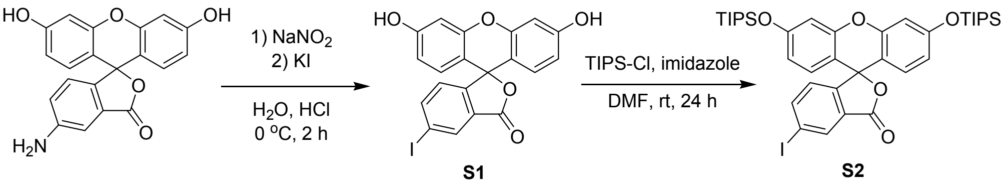


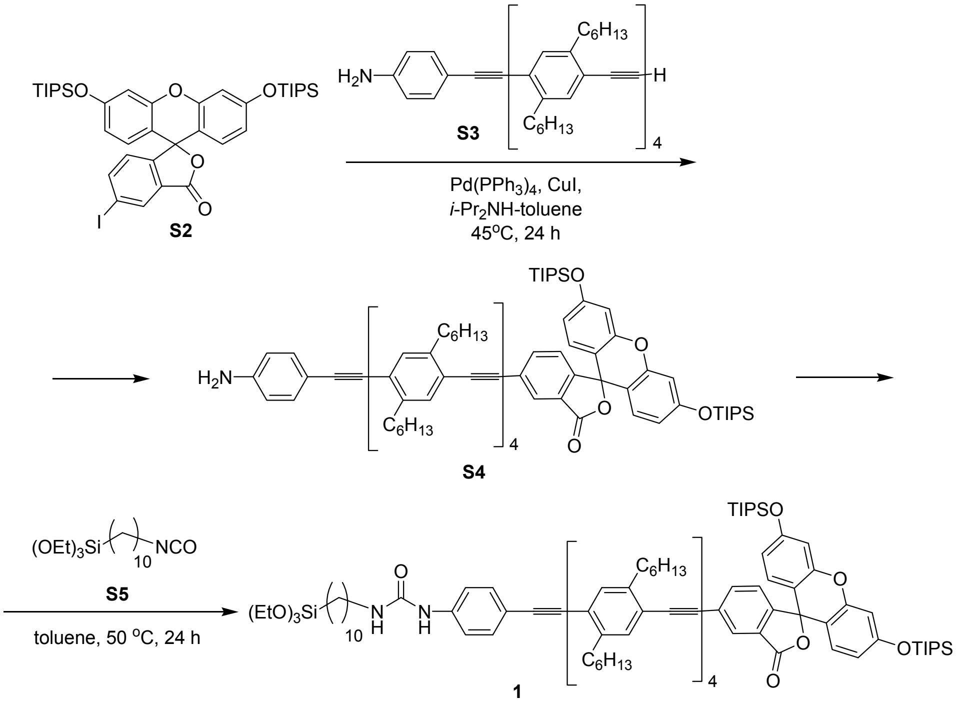
Figure S8. Cyclic voltammogram of a monolayer of **1** immobilized on ITO/glass electrode surface. Experimental conditions: 0.1 M Bu_4NPF_6 in CH_2Cl_2, sweep rate 0.1 V s^{-1}.

Synthetic Details



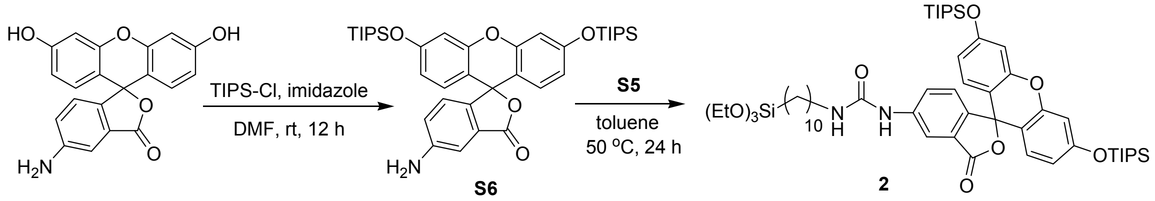
5-Iodofluorescein (S1) was synthesized according to literature procedure.[4]

Bis(triisopropylsilyl)-5-iodofluorescein (S2). To a solution of 0.34 g (0.74 mmol) of **S1** and 0.36 g (5.30 mmol) of imidazole in 5 ml of anhydrous DMF, 0.50 g (0.55 ml, 2.60 mmol) of neat triisopropylsilyl chloride (TIPS-Cl) was added dropwise at room temperature. The reaction mixture was stirred at room temperature for 24 h, then poured into CH_2Cl_2 and washed successively with water, brine, and dried over $MgSO_4$. After concentration in vacuo, the crude product was purified by column chromatography on silica gel (eluent hexanes – EtOAc 9:1) yielding 0.46 g (59%) of **S2** as a colorless oil, R_f 0.35. ^{1}H NMR ($CDCl_3$) δ 8.34 (d, J = 1.5 Hz, 1H); 7.95 (dd, , J_1 = 8.0, J_2 = 1.5 Hz, 1H); 6.93 (d, , J = 8.0 Hz, 1H); 6.75 (d, J = 2.0 Hz, 2H); 6.65-6.50 (m, 4H); 1.28 (septet, J = 7.5 Hz, 6H); 1.08 (d, J = 7.5 Hz, 36H). HRMS (ESI-TOF) *m/e* 771.2385 M$^+$ (calcd. for $C_{38}H_{52}IO_5Si_2$ 771.2392).



Compound S4. A mixture of 0.48 g (0.45 mmol) of alkyne **S3** (prepared following the literature procedure[1]), 0.38 g (0.49 mmol) of iodide **S2**, 2 mg (2 μmol) of $Pd(PPh_3)_4$ and one grain of CuI in 3 ml of toluene-diisopropylamine (7:3) was stirred in a sealed Schlenk flask at 45 °C for 24 h. After allowing to cool to room temperature, the reaction mixture was poured into CH_2Cl_2 and washed successively with saturated $NaHCO_3$, water, and brine. After concentration in vacuo, the crude product was further purified by column chromatography on silica gel (eluent CH_2Cl_2 – hexanes 1:1) to yield 0.26 g (31%) of **S4** as a yellow oil, R_f 0.40. ^{1}H NMR ($CDCl_3$) δ 8.16 (s, 1H); 7.79 (d, *J* = 8.5 Hz, 1H); 7.71 (broad s, 1H); 7.50-7.30 (m, 10H); 7.18 (d, *J* = 7.8 Hz, 2H); 6.78 (s, 2H); 6.70-6.50 (m, 4H); 3.87 (broad s, 2H); 2.92-2.72 (m, 16H); 1.80-1.65 (m, 16H); 1.55-1.20 (m, 50H); 1.20-1.05 (m, 36H); 1.00-0.80 (m, 24H).

Fluoride sensor compound 1. A solution of 76 mg (41 μmol) of compound **S4** and 14 mg (41 μmol) of triethoxy-(10-isocyanatodecyl)silane **S5** (prepared following the literature procedure[5]) in 1 ml of toluene was stirred at 50 °C for 24 h. After concentration in vacuo, the crude product was purified by column chromatography on silica gel (eluent CH_2Cl_2 – hexanes 1:2) to yield **1** in quantitative yield as a brown oil, R_f 0.50. ^{1}H NMR ($CDCl_3$) δ 8.12 (s, 1H); 7.85-7.75 (m, 2H); 7.50-7.30 (m, 10H); 7.15 (d, *J* = 2.5 Hz, 2H); 6.75 (d, *J* =2.4 Hz, 2H); 6.66 (d, *J* = 8.8 Hz, 2H); 6.57 (dd, J_1 = 8.8, J_2 = 2.4 Hz, 2H); 6.24 (s, 1H); 4.60-4.50 (m, 1H); 3.81 (q, *J* = 7.0 Hz, 6H); 3.30-3.20 (m, 2H); 2.90-2.70 (m, 16H); 1.82-1.62 (m, 16H); 1.50-1.20 (m, 79H); 1.10 (d, *J* = 5 Hz, 36H); 0.90-0.80 (m, 24H); 0.70-0.60 (m, 2H). MS (MALDI-TOF) *m/e* M^+ 2177.49 (calcd. for $C_{143}H_{204}N_2O_9Si_3$ 2178.54).



Bis(triisopropylsilyl)-5-aminofluorescein S6. Triisopropylsilylchloride (0.27 g, 0.3 ml, 1.44 mmol) was added dropwise to a solution of 100 mg (0.29 mmol) of 5-aminofluorescein and 100 mg (1.47 mmol) of imidazole in 0.5 ml of anhydrous DMF, and the reaction mixture was stirred for 12 h at room temperature. Hexane (30 ml) and ethyl acetate (15 ml) were added to the reaction mixture followed by washing with water (3×15 ml), brine (15 ml), and drying over Na_2SO_4. After concentration in vacuo, the crude white solid was purified by washing with cold hexane and drying under vacuum, yielding 135 mg (71%) of **S6** as a pale yellow solid, mp. 279-280 °C (decomp.). ^1H NMR ($CDCl_3$) δ 7.18 (s, 1H), 6.93 (s, 2H), 6.80-6.60 (m, 4H), 6.55 (d, *J* = 7.5 Hz, 2H), 1.45-1.10 (m, 6H), 1.06 (d, *J* = 8.3 Hz, 36H). HRMS (ESI-TOF) *m/e* M^+ 660.3534 (calcd. for $C_{38}H_{54}NO_5Si_2$ 660.3535).

Reference compound 2. A solution of 54 mg (82 μmol) of **S6** and 28 mg (81 μmol) of isocyanate **S5** in 1 ml of toluene was stirred at 50 °C for 24 h, and then was diluted with 135 ml of toluene to make a ~0.6 mM stock solution of **2** required for its immobilization on glass. A small portion of the reaction mixture was concentrated in vacuo for NMR characterization. ^1H NMR ($CDCl_3$) δ 8.33 (d, *J* = 7.0 Hz, 1H), 8.18 (s, 1H), 7.79 (s, 1H), 7.10 (d, *J* = 7.0 Hz, 1H), 6.80-6.50 (m, 6H), 5.45 (broad s, 1H), 3.81 (q, *J* = 7.0 Hz, 6H), 3.20-3.00 (m, 2H), 1.50-1.00 (m, 67H); 0.70-0.60 (m, 2H).

References

[1] Imsick, B. G.; Acharya, J. R.; Nesterov, E. E. *Adv. Mater.* **2012,** *24,* 120-124.

[2] Osaheni, J. A.; Jenekhe, S. A. *J. Am. Chem. Soc.* **1995**, *117*, 7389-7398.

[3] Martínez, V. M.; Arbeloa, F. L.; Prieto, J. B.; Arbeloa, I. L. *Chem. Mater.* **2005**, *17*, 4134-4141.

[4] Jiao, G.-S.; Han, J. W.; Burgess, K. *J. Org. Chem.* **2003**, *68*, 8264-8267.

[5] Joly, P.; Ardès-Guisot, N.; Kar, S.; Granier, M.; Durand, J.-O.; Melnyk, O. *Eur. J. Org. Chem.* **2005**, 2473-2480.

^{1}H NMR Spectra of selected compounds

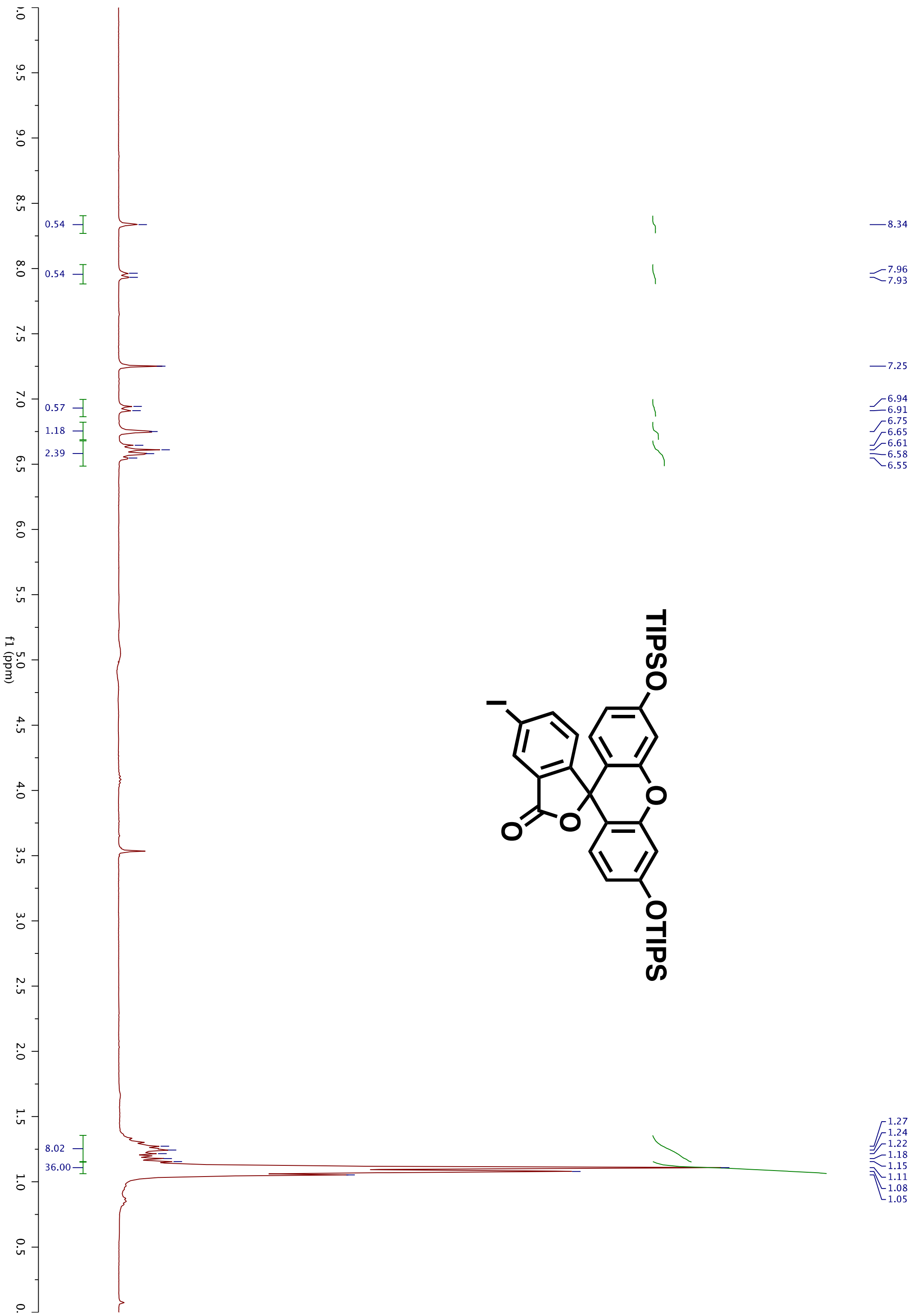


Figure S9. ^{1}H NMR spectrum of compound **S2** ($CDCl_3$, 250 MHz).

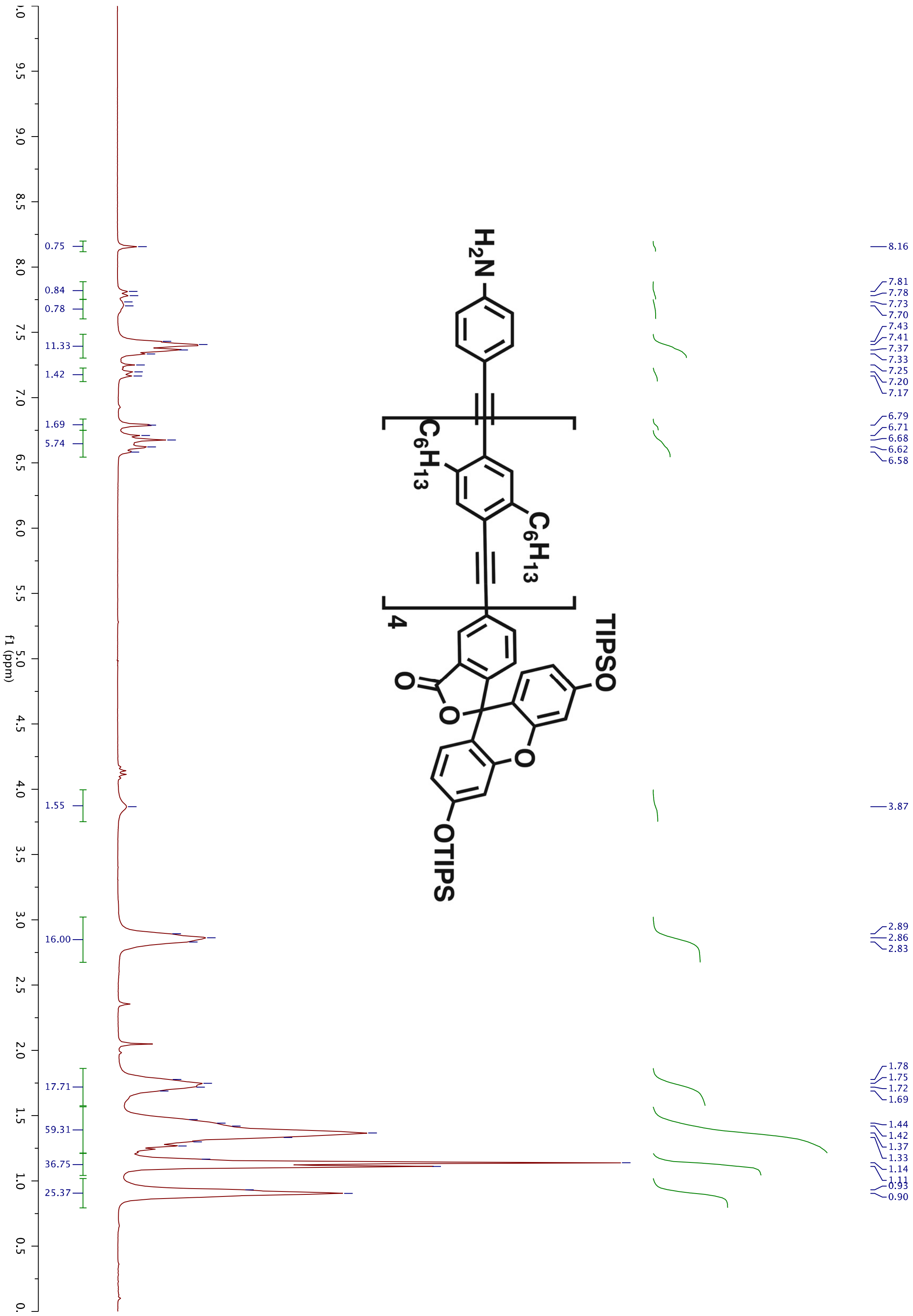


Figure S10. ^{1}H NMR spectrum of compound **S4** ($CDCl_3$, 250 MHz).

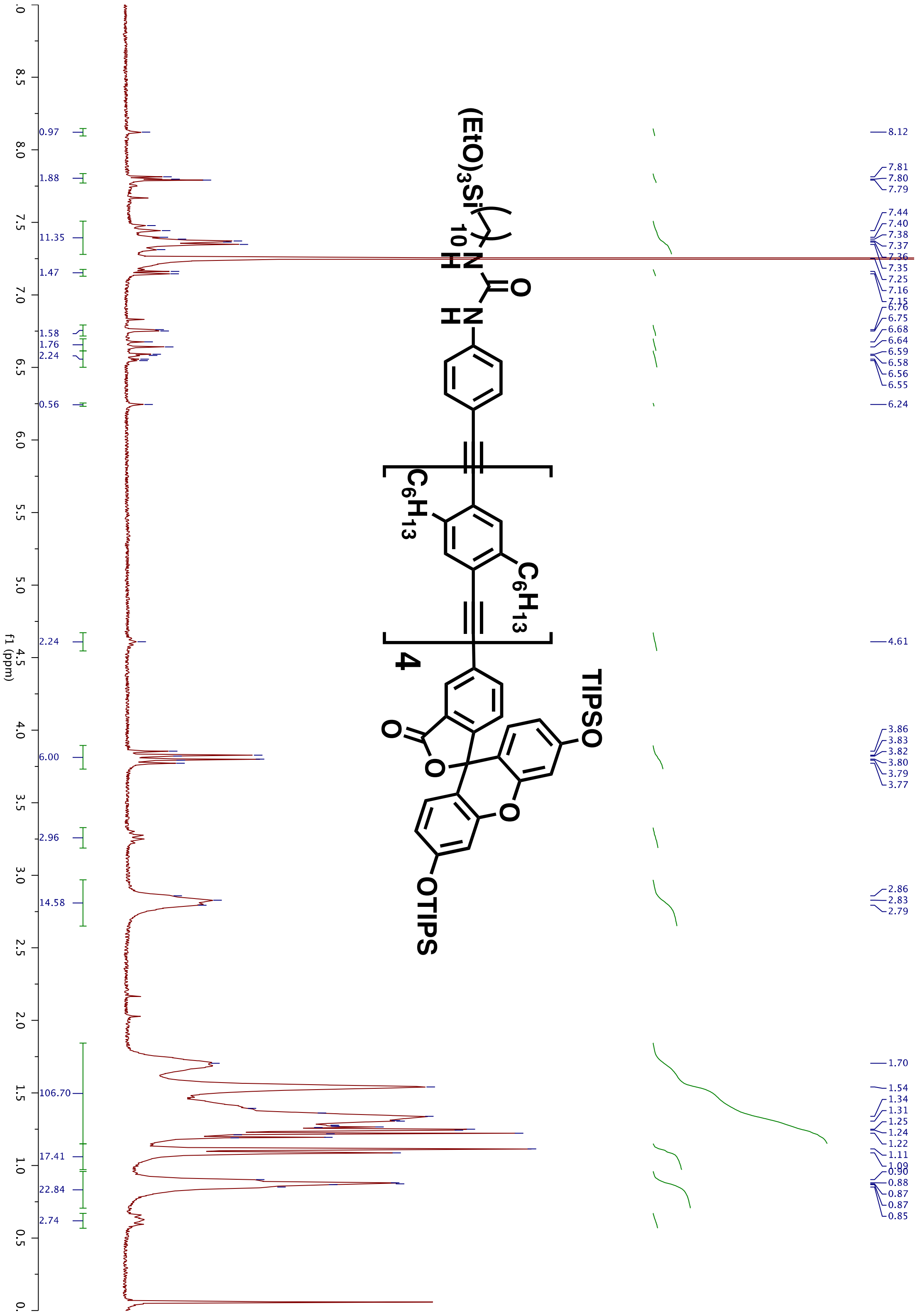


Figure S11. ^{1}H NMR spectrum of compound **1** ($CDCl_3$, 250 MHz).

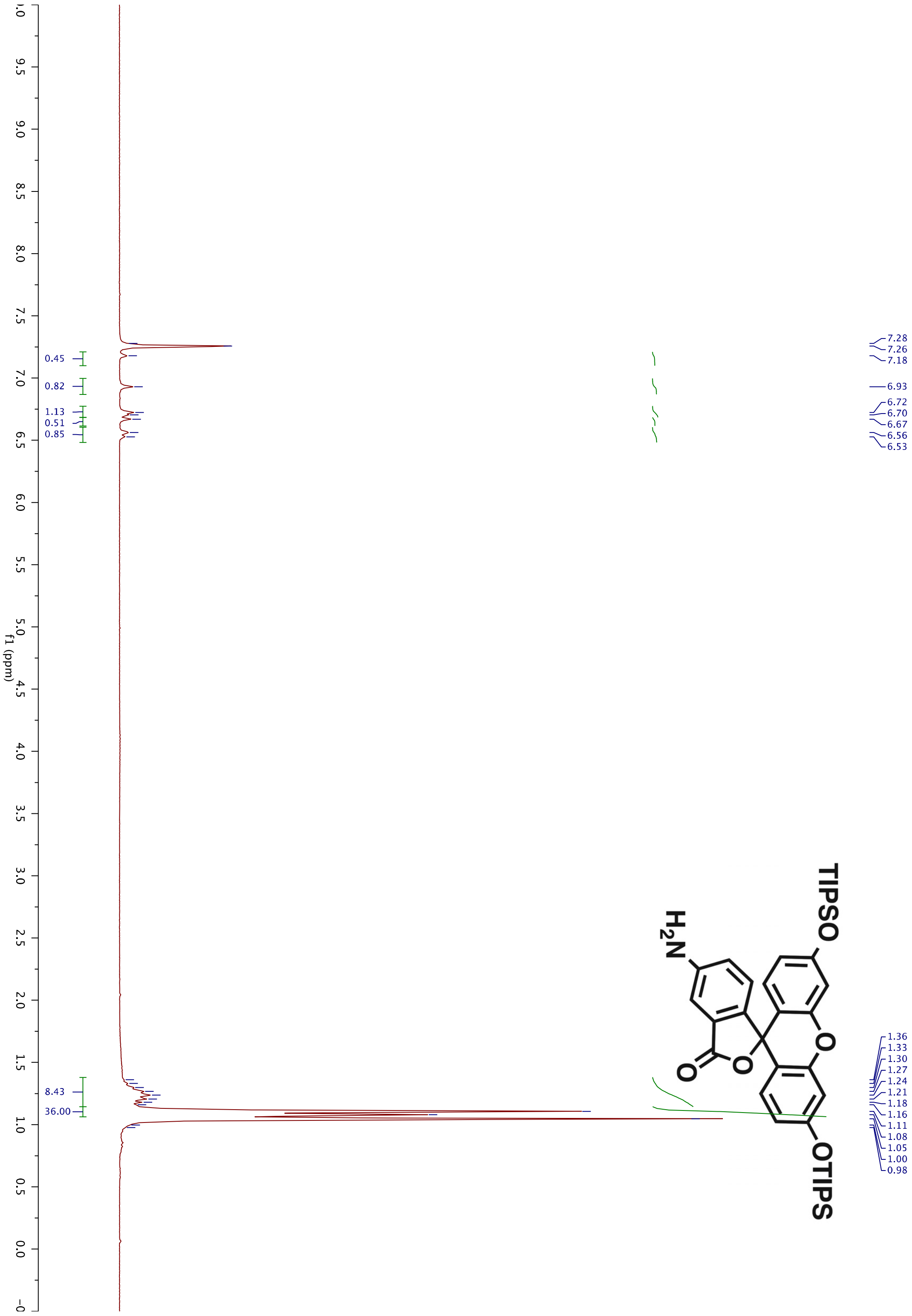


Figure S12. ^{1}H NMR spectrum of compound **S6** (CDCl$_3$, 250 MHz).

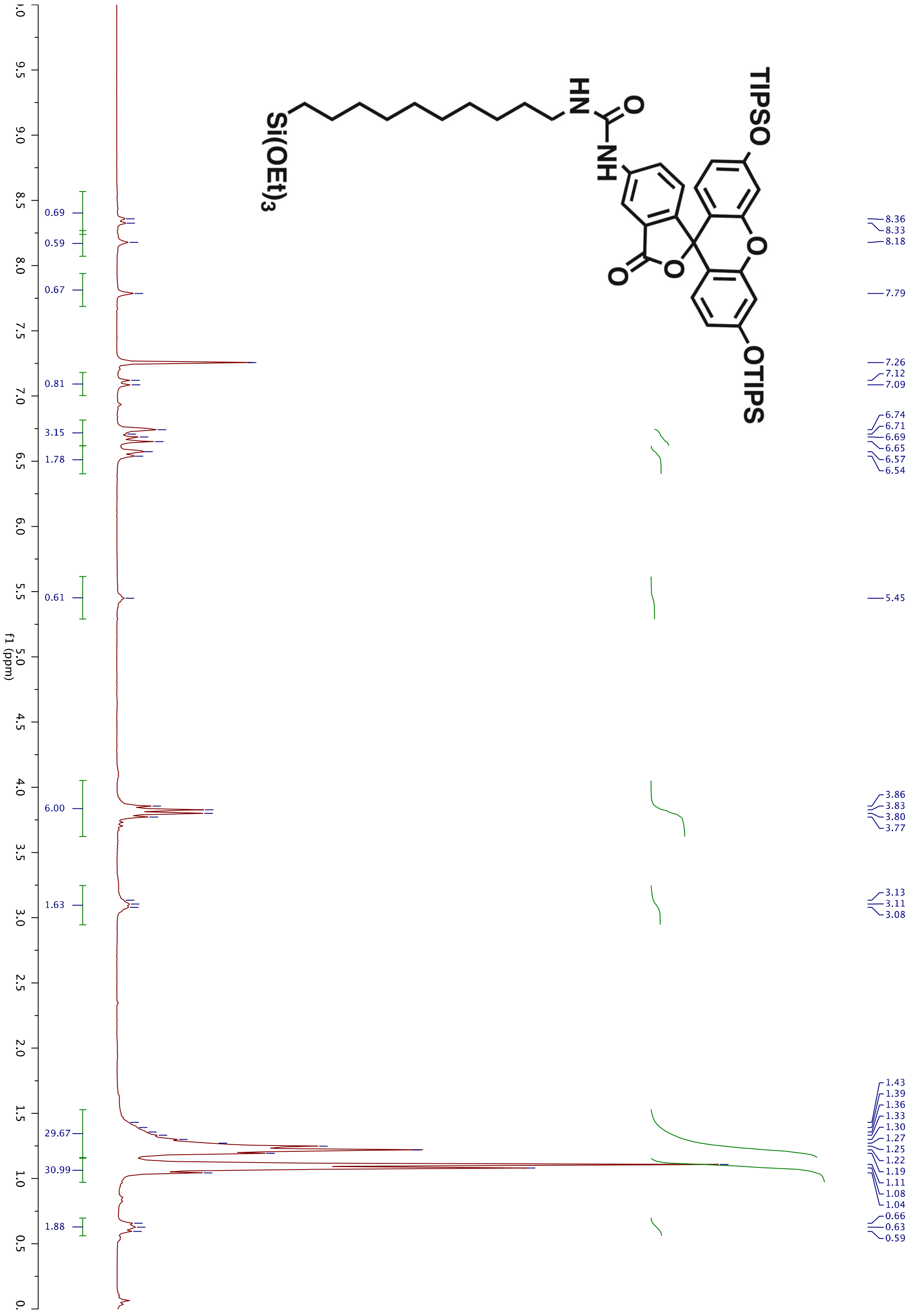


Figure S13. ^{1}H NMR spectrum of compound **2** ($CDCl_3$, 250 MHz).