Supporting information

From Slow to Fast – the User Controls the Rate of the Release of Molecules From Masked Forms Using a Photoswitch and Different Types of Light

*C. Chad Warford, Carl-Johan Carling and Neil R. Branda**

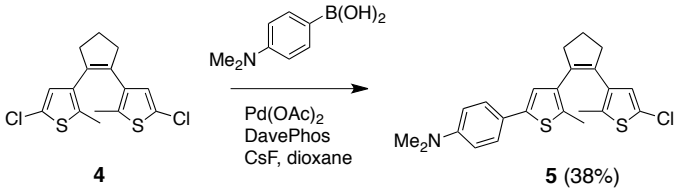
Materials and Methods

General. All solvents and reagents used for synthesis, chromatography, UV–vis spectroscopy and photolysis studies were purchased from Aldrich with the exception of anhydrous DMF, which was purchased from Acros. Unless otherwise noted, all solvents were dried and degassed by passing them through steel columns containing activated alumina under nitrogen using an MBraun solvent purification system. Solvents for NMR analysis were purchased from Cambridge Isotope Laboratories and used as received. Column chromatography was performed using silica gel 60 (230–400 mesh) from Silicycle Inc. *n*-Butyllithium and *tert*-butyllithum were titrated against diphenylacetic acid. The dichloride starting material **4** and was prepared according to the literature procedure.[1]

Instrumentation. ^{1}H, ^{13}C, ^{19}F, and ^{31}P NMR characterizations were performed on a Bruker AMX 400 instrument with a 5 mm inverse probe operating at 400.13 MHz for ^{1}H NMR, 471 MHz for ^{19}F NMR, 243 MHz for ^{31}P NMR and 100.6 MHz for ^{13}C NMR, a Bruker 500 operating at 500.13 MHz for ^{1}H NMR, and 125.75 MHz for ^{13}C NMR, or a Bruker Avance II 600 with a 5 mm QNP cryoprobe operating at 600.45 MHz for ^{1}H NMR and 150.90 MHz for ^{13}C NMR. Chemical shifts (δ) are reported in parts per million relative to tetramethylsilane using the residual solvent peak as a reference. Coupling constants (J) are reported in Hertz. IR spectra were recorded on a Thermo Nicolet Nexus 670 FTIR spectrometer equipped with a Pike MIRacle ATR sampling accessory. High Resolution Mass Spectroscopy (HRMS) measurements were performed using an Agilent 6210 TOF LC/MS in ESI-(+) mode. Melting Points were measured using a Fisher-Johns melting point apparatus, or a Gallenkamp melting point apparatus (Registered Design No. 889339).

Photoinduced ring-closing and photolysis reactions. Standard hand-held lamps used for visualizing TLC plates were used to carry out the ring-closing and photolysis reactions at 365 nm (1.4 mW/cm^2, 3.9 mW). Visible light irradiation was carried out using the light of a 150-W tungsten source that was passed through a 434-nm cutoff filter to eliminate higher-energy light. Irradiation was carried out in low-light conditions to minimize interference from ambient light. Irradiation with low-intensity monochromatic light was generated using the source of a PTI Quantamaster Spectrofluorimeter with a 2 nm slit width.

[1] L. N. Lucas, J. van Esch, R. M. Kellogg, B. L. Feringa, *Chem. Commun.* **1998**, *21*, 2313–2314.

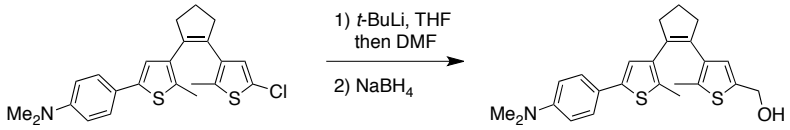


4 **5** (38%)

Synthesis of 1-(2-chloro-5-methylthien-3-yl)-2-(2-(4-(dimethylamino)phenyl)-5-methylthien-3-yl)cyclopentene (5). A flame-dry 3-neck flask containing a stir bar and equipped with a flame-dried reflux condenser was charged with dichloride **4** (667 mg, 2.02 mmol), CsF (923 mg 6.07 mmol), 4-*N,N*-dimethylaminophenylboronic acid (401 mg, 2.43 mmol) and $Pd(OAc)_2$ (23 mg, 0.10 mmol), then transferred to a glove box, where 2-dicyclohexylphosphino-2'-(*N,N*-dimethylamino)biphenyl (Davephos, 60 mg, 0.15 mmol) was added. The flask was sealed with a rubber septum, removed from the glove box, and immediately connected to a nitrogen manifold under positive N_2 pressure via a needle. The flask was charged with dry 1,4-dioxane (35 mL) via a cannula resulting in a deep red solution containing a suspended white powder. The reaction mixture was stirred at room temperature for 15 h, at which time it was quenched by pouring it into water (35 mL). The layers were separated and the aqueous layer was extracted with Et_2O (3 × 20 mL). The combined organic layers were washed with water (50 mL), then brine (50 mL), dried over anhydrous Na_2SO_4, filtered, and concentrated under vacuum to afford a yellow-brown oil. Purification by flash chromatography under red light (10% EtOAc in hexanes) yielded 315 mg (38%) of compound **5** as an off-white solid.

^{1}H NMR (400 MHz, CD_2Cl_2) δ 7.35 (d, J = 8.9 Hz, 2H), 6.83 (s, 1H), 6.69 (d, J = 8.9 Hz, 2H), 6.64 (s, 1H), 2.95 (s, 6H), 2.84-2.77 (m, 2H), 2.74 (m, 2H), 2.10-2.00 (m, 2H), 1.97 (s, 3H), 1.91 (s, 3H).

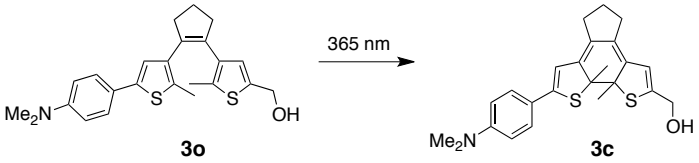
^{13}C NMR (101 MHz, $CDCl_3$) δ 149.75, 140.56, 136.03, 135.62, 135.24, 133.28, 133.25, 132.32, 126.87, 126.28, 126.26, 124.80, 121.56, 112.62, 40.56, 38.45, 38.34, 22.91, 14.29, 14.17.



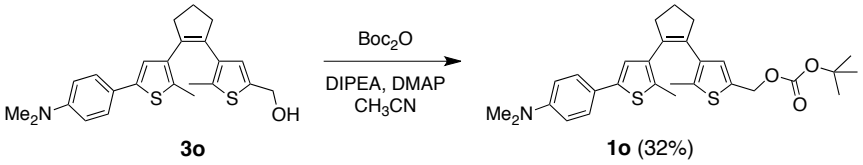
5 **3o** (81%)

Synthesis of 1-(2-hydroxymethyl-5-methylthien-3-yl)-2-(2-(4-(dimethylamino)-phenyl)-5-methylthien-3-yl)cyclopentene (3o). A solution of **5** (68 mg, 0.16 mmol) in dry THF (25 mL) was wrapped in aluminum foil to avoid photochemistry, cooled to -78 °C in a dry ice/acetone bath under a nitrogen atmosphere and treated with *tert*-butyllithium (200 µL, 1.64 M, 0.328 mmol) dropwise via cannula through a rubber septum. After stirring for 15 min, the starting material had been consumed as monitored by TLC (hexanes). The reaction mixture was treated with DMF (26 µL, 0.34 mmol) using a syringe and stirred for 15 min, at which time the cooling bath was removed and the reaction was allowed to warm to room temperature over 5 h. The reaction mixture was concentrated under vacuum to afford yellow oil, which was dissolved in a 1:1 mixture of EtOH and THF (35 mL). The resulting yellow solution was cooled by immersion in an ice-water bath at 0 °C and treated with solid $NaBH_4$ (6.8 mg, 180 mmol) under a flow of

nitrogen. The reaction mixture was allowed to warm to room temperature over 18 h, at which time it was quenched with a saturated aqueous solution of ammonium chloride. The aqueous layer was separated and washed with EtOAc (3 × 50 mL). The combined organic layers were washed with water (25 mL), then brine (50 mL), dried over anhydrous Na_2SO_4, filtered, and concentrated under vacuum to afford a pale yellow oil. Purification by flash chromatography under red light (5% EtOAc in hexanes) yielded 54 mg (81%) of compound **3o** as an off-white solid.
^{1}H NMR (400 MHz, CD_3CN) δ 7.38-7.30 (m, 2H), 6.88 (s, 1H), 6.75-6.69 (m, 2H), 6.67 (s, 1H), 4.54 (d, J = 4.4 Hz, 2H), 3.19 (br s, 1H), 2.92 (s, 6H), 2.77 (dt, J = 15.3, 7.6 Hz, 4H), 2.03 (m, 2H), 1.969 (s, 3H), 1.949 (s, 3H).
^{13}C NMR (101 MHz, CD_2Cl_2) δ 150.44, 140.84, 140.39, 137.08, 136.10, 135.41, 134.91, 132.67, 127.17, 126.60, 123.41, 122.24, 113.03, 60.52, 40.84, 39.02, 39.01, 23.55, 14.70, 14.55.
HRMS (ESI-(+)) Anal. Calc. 410.1607, found 410.1607.
UV-vis (CH_3CN, 20.4 μM) λ_{max} = 328 nm, ε_{328} = 24800.

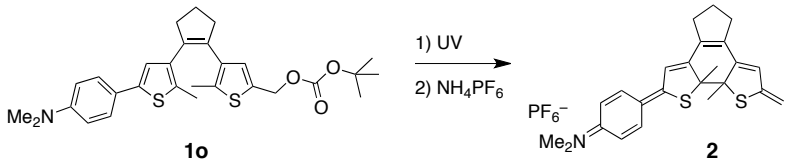


Ring-closing of 1-(2-hydroxymethyl-5-methylthien-3-yl)-2-(2-(4-(dimethylamino)-phenyl)-5-methylthien-3-yl)cyclopentene (3o). A CH_3CN solution of **3o** was irradiated with 365 nm light until all changes in the UV-vis spectra stopped. The red solution contained 82% of the ring-closed isomer as measured by ^{1}H NMR spectroscopy.
UV-vis (CH_3CN, 20.4 μM) λ_{max} = 495 nm, ε_{495} = 21100 (based on an 82% photostationary state).



Synthesis of 1-(2-butoxycarbonyloxymethyl-5-methylthien-3-yl)-2-(2-(*N,N*-dimethylanilin-4-yl)-5-methylthien-3-yl)cyclopentene (1o). A clear, colourless solution of **3o** (122 mg, 0.297 mmol) in dry CH_3CN (50 mL) in a 3-neck flask was treated with DMAP (7.2 mg, 59 μmol) and di-*tert*-butyl dicarbonate (98 mg, 0.45 mmol) under a nitrogen flow to form a clear, colourless solution. The flask was sealed and cooled by immersion in an ice-water bath at 0 °C. After stirring for 5 min to stabilize the temperature, the reaction was treated with *N,N*-di-isopropylethylamine (78 μL, 0.45 mmol) drop wise using a syringe. The resulting clear, colourless solution was wrapped in aluminum foil and stirred for 25 min, at which time the cooling bath was removed and the reaction mixture stirred for 18 h. The reaction mixture was quenched by pouring it into 1M HCl (25 mL). The aqueous layer was separated and extracted with EtOAc (3 × 25 mL). The combined organic layers were washed with a saturated aqueous solution of sodium bicarbonate (25 mL), then water (25 mL), then brine (15 mL), dried over Na_2SO_4,

filtered and evaporated to dryness under vacuum. Purification by flash chromatography under red light (5% EtOAc in hexanes) yielded 48 mg (32%) of compound **1o** as an off-white solid.
^{1}H NMR (400 MHz, $CDCl_3$) δ 7.37 (d, J = 8.9 Hz, 2H), 6.84 (s, 1H), 6.82 (s, 1H) 6.71 (d, J = 8.9 Hz, 2H), 5.10 (s, 2H), 2.97 (s, 6H), 2.79 (m, 4H), 2.11-1.99 (m, 2H), 1.94 (s, 3H), 1.91 (s, 3H), 1.49 (s, 9H).
^{13}C NMR (101 MHz, CD_2Cl_2) δ 153.77, 150.45, 140.93, 137.02, 136.19, 135.65, 134.64, 133.90, 132.69, 130.12, 126.61, 122.15, 113.02, 82.63, 63.65, 40.83, 39.03, 28.06, 23.53, 14.66, 14.52.
HRMS (ESI-(+)) Anal Calc. 510.2131, found 510.2125.
UV-vis (CH_3CN, 20.04 μM) λ_{max} = 329 nm, ε_{329} = 39600.



1o **2**

Synthesis of 2-(4-(*N,N*-dimethylamino)phenyl)-9a,9b-dimethyl-8-methylene-4,5,6,8,9a,9b-hexahydroindeno[5,4-b:6,7-b']dithiophen-1-ium hexafluorophosphate (2•PF_6). A quartz photolysis chamber under nitrogen atmosphere was charged with **1o** (35 mg, 69 μmol) and purged with nitrogen for 1 h. The reaction chamber was then charged with 5% v/v degassed water/CH_3CN mixture (300 mL) and stirred to form a clear, colourless solution. The apparatus was wrapped in aluminum foil and partially submerged in an ice-water bath and left to equilibrate for 5 min. The solution was irradiated using a Hanovia 697-A36 mercury arc lamp (450 W) for 70 min, at which time TLC analysis (20 % EtOAc in hexanes) indicated no starting material remained. The deep purple reaction mixture was concentrated under vacuum to approximately 10 mL, then added drop wise into a vigorously stirred aqueous solution (15 mL) of NH_4PF_6 (186 mg, 1.14 mmol). A blue-black precipitate formed, which was isolated by centrifugation, then washed by re-suspending it in water (triplicate), then hexanes (triplicate). The resulting solid was dissolved in CH_2Cl_2, evaporated under vacuum to a solid residue then dried under vacuum overnight. The purple solid was dissolved in CH_2Cl_2 and again added drop wise into vigorously stirred hexanes (40 mL). The resulting fine purple precipitate was then stirred gently overnight and isolated by centrifugation with triplicate re-suspension in hexanes to yield 7.2 mg (20%) of **2•PF_6**.
HRMS (ESI-(+)): 392.1486 Anal. Calcd. 392.1501.
UV-vis (CH_3CN, 59.5 μM) λ_{max} = 571 nm, ε_{571} = 3400.

Kinetic experiment to determine the rate of release from 1o. A solution of a 15% v/v mixture of 105 mM tris buffer at pH 6.5 with a CH_3CN solution of **1o** (20 μM) in a cuvette was exposed to 365 nm light for 70 s to generate approximately 80% of **1c**, then left in the dark for 50 h. An absorbance reading at 570 nm was taken automatically every 10 min. The absorbance vs. time data were fit using GraphPad Prism to the following equation:

$$1c \overset{k_1}{\rightarrow} 2 \overset{k_2}{\rightarrow}$$

$$[\mathbf{2}] = a_0 \left(\frac{k_1}{k_1 - k_2}\right)(e^{-k_2 t} - e^{-k_1 t}) + x$$

where the concentration of cation **2** is assumed to be proportional to the absorbance, a_0 is the initial concentration of **1c**, k_1 is the rate of formation of **2**, k_2 is the rate of disappearance of **2**, and x is a parameter to account for the initial non-zero concentration of **2** at time = 0 due to some conversion of **1o** during the initial exposure to 365 nm and t is time. The results of the regression produced values for k_1 and k_2 of $(2.7 \pm 0.02) \times 10^{-3}$ s^{-1} and $(5.3 \pm 0.02) \times 10^{-4}$ s^{-1}, respectively (R = 0.9986).

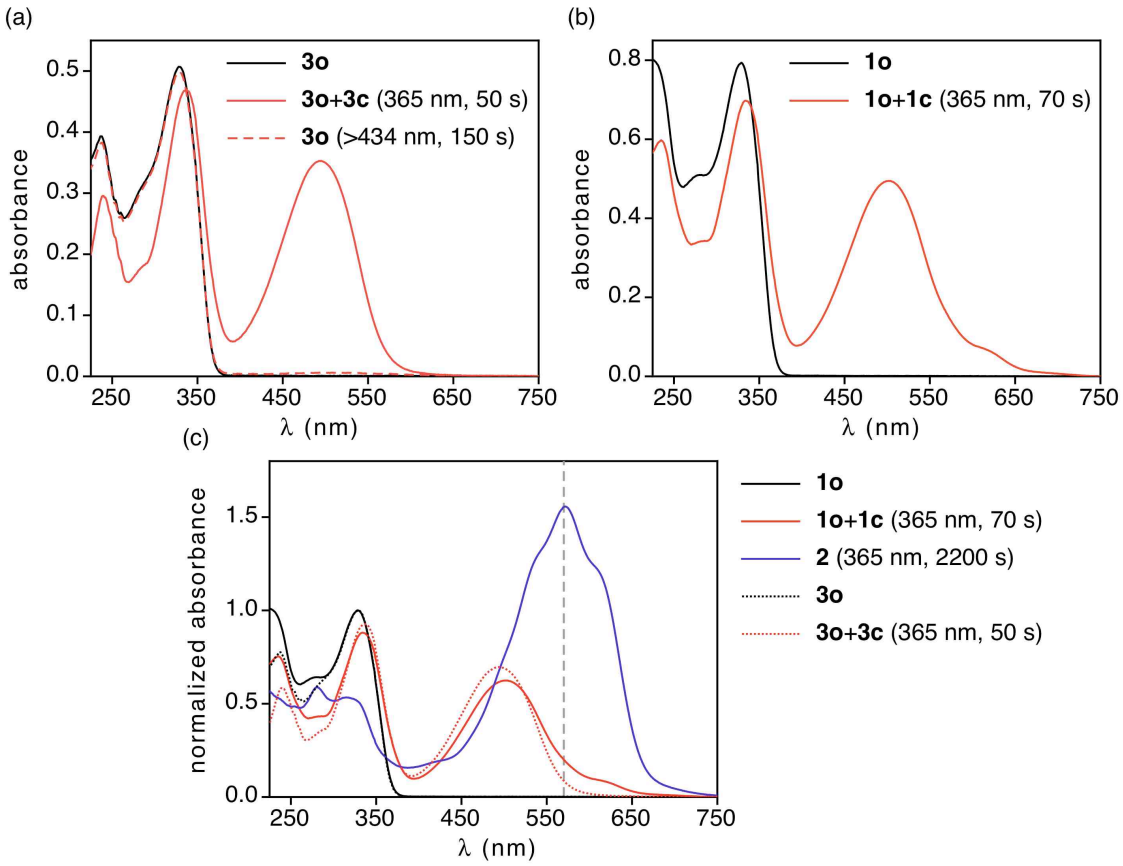


Figure S1. (a) UV-vis absorption spectra of a CH_3CN solution of alcohol **3o** (2×10^{-5} M) before (black line) and after exposure to 365 nm light for 50 s (red solid line), and after exposure to >434 nm light for 150 s (red dashed line). (b) UV-vis absorption spectra of a wet CH_3CN solution (5% v/v H_2O) of carbonate **1o** (20 μM) before (black line) and after exposure to 365 nm light for 70 s (red solid line). (c) UV-vis absorption spectra of the photostationary state of **3o/c**, **1o/c** and after **1o/c** is exposed to 365 nm for 2200 s to produce cationic, quinoid **2**.

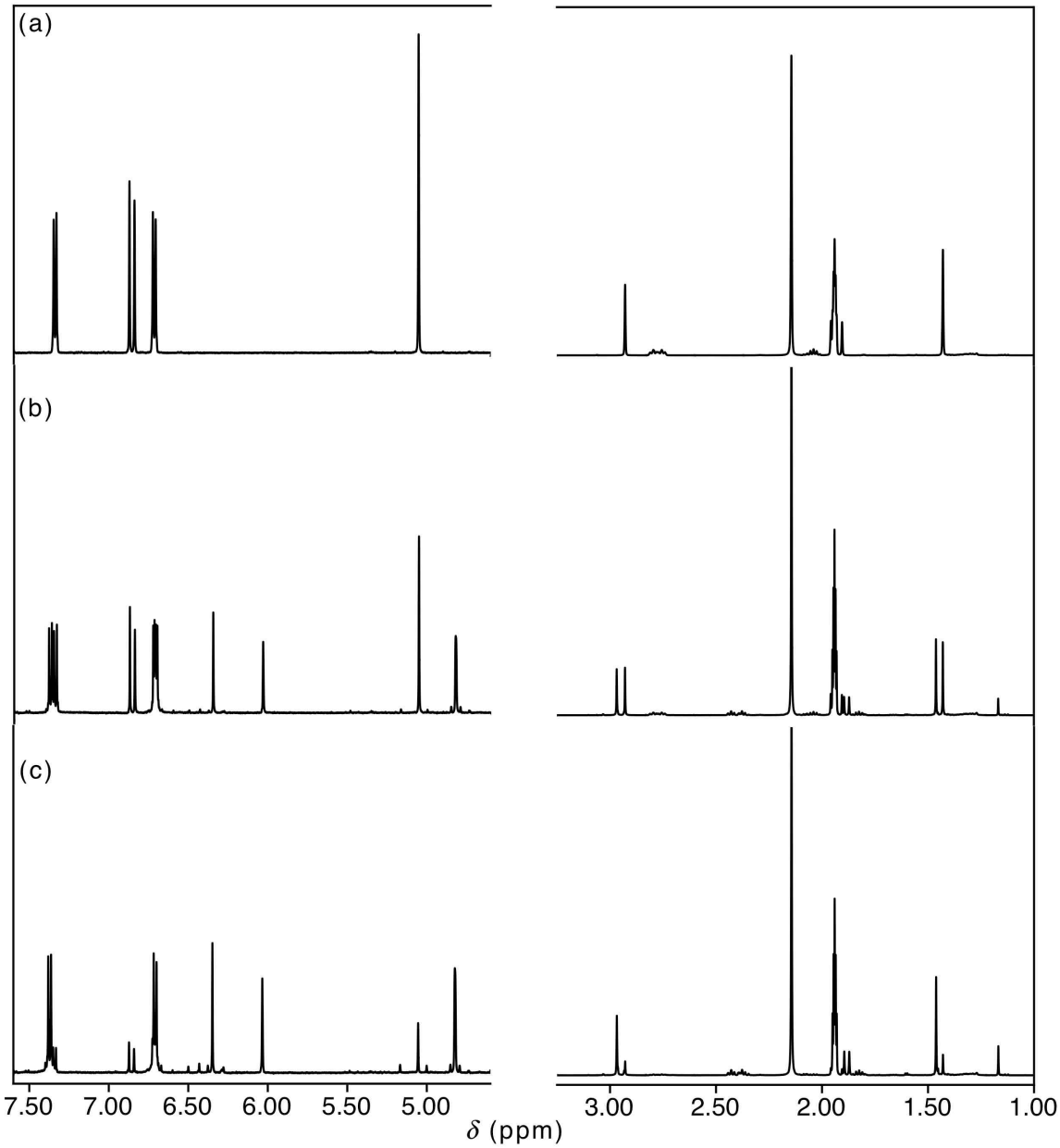


Figure S2. ^{1}H NMR spectra (CD_3CN/400 MHz) of carbonate **1o** (a) before, (b) during, and (c) after exposure to 365 nm light for 700 s. The intermediate spectrum is shown to clearly illustrate the signals that are changing as the ring-open isomer is converted into the ring-closed isomer.

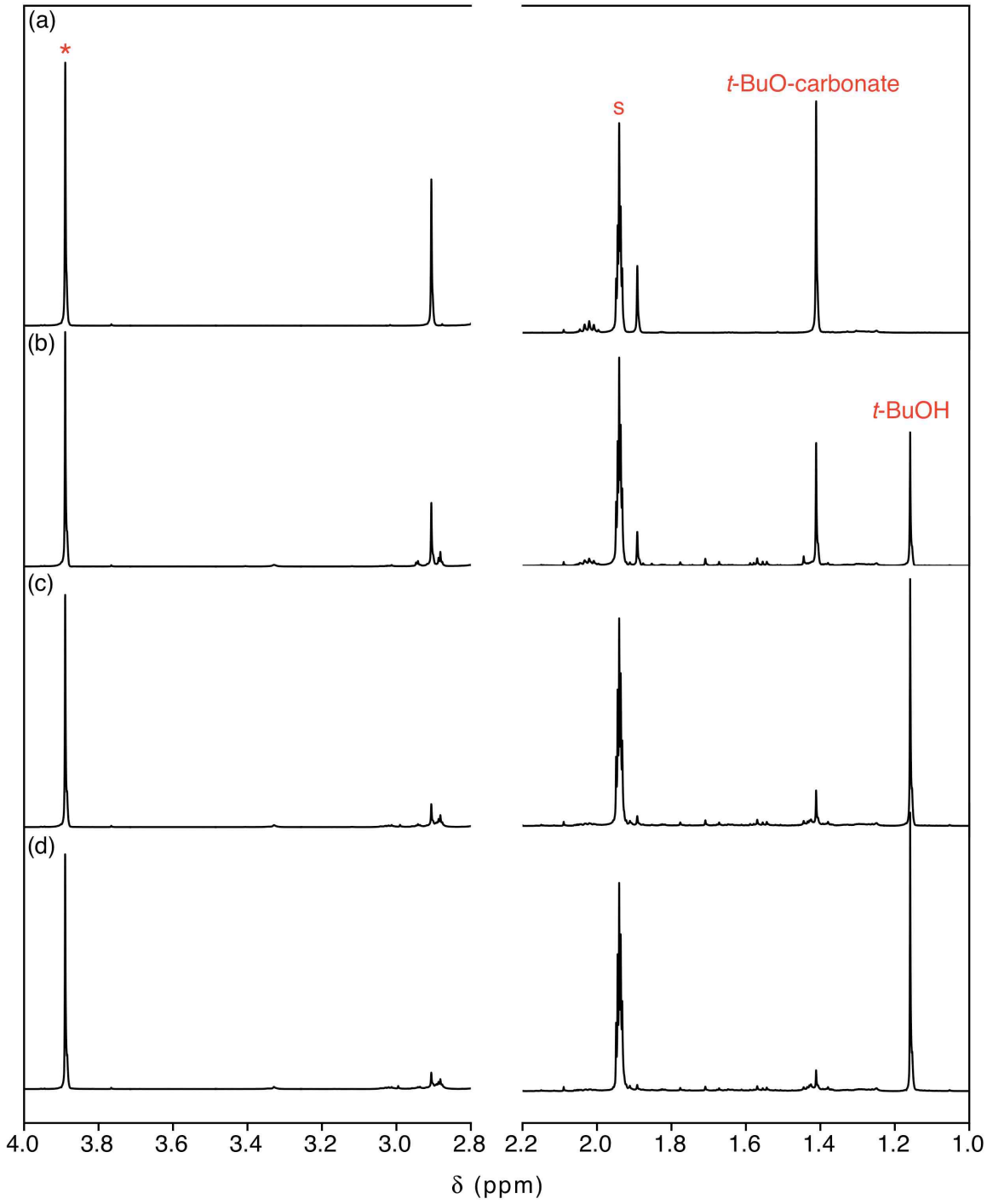


Figure S3. Partial ^{1}H NMR spectra (5 v/v % D_2O/CD_3CN, 600 MHz) of a mixture of carbonate **1o** (6.65 mM) and *p*-nitroanisole (17.5 mM, identified with the red asterisk*) after simultaneous exposure to 365 nm and ≥ 434 nm irradiation for 120 min.

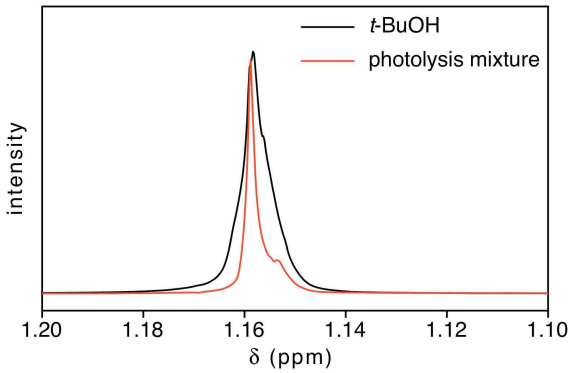


Figure S4. ^{1}H NMR spectra (5 v/v % D_2O/CD_3CN, 600 MHz) of the photolysis reaction mixture of carbonate **1o** (6.65 mM) after simultaneous exposure to 365 nm and ≥ 434 nm irradiation for 120 min (red trace) and *tert*-butanol (black trace).

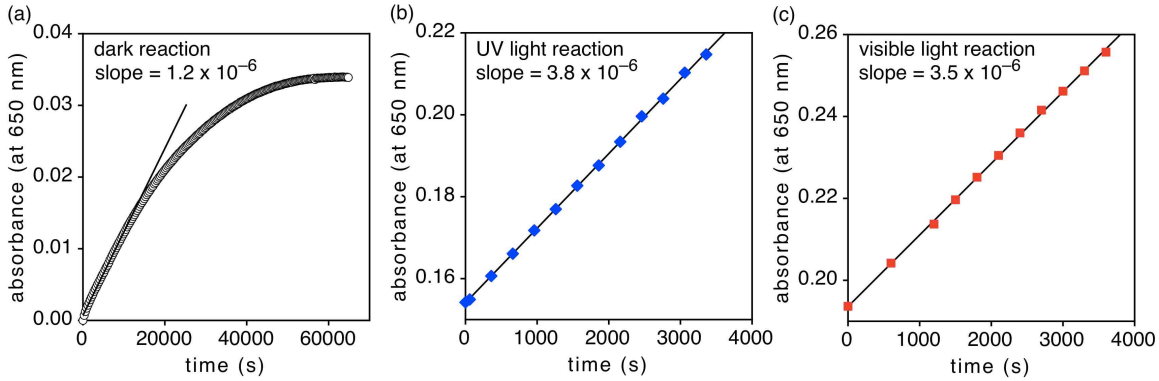


Figure S5. Changes in the absorbances at 650 nm that correspond to compound **2** when solutions are irradiated with 365 nm light for 70 s and then (a) left in the dark, (b) exposed to continuous monochromatic 342 nm light, and (c) exposed to monochromatic 485 nm light. The lines show the initial slopes.

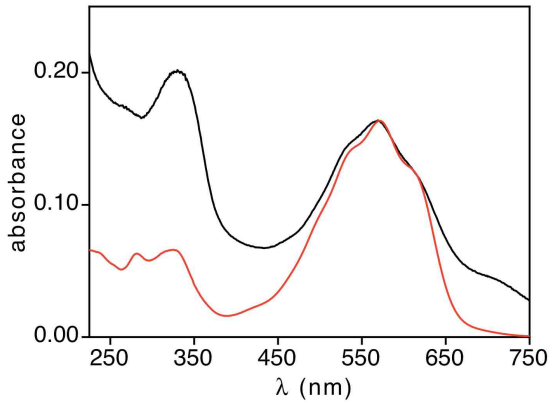


Figure S6. Normalized UV-vis absorption spectra of a CH_3CN solution of **2**·PF_6 isolated from the bulk photolysis experiment (black line) and the spectrum that results when a wet CH_3CN solution (5% v/v H_2O) of **1o** is irradiated with 365 nm light for 2200 s (red line).

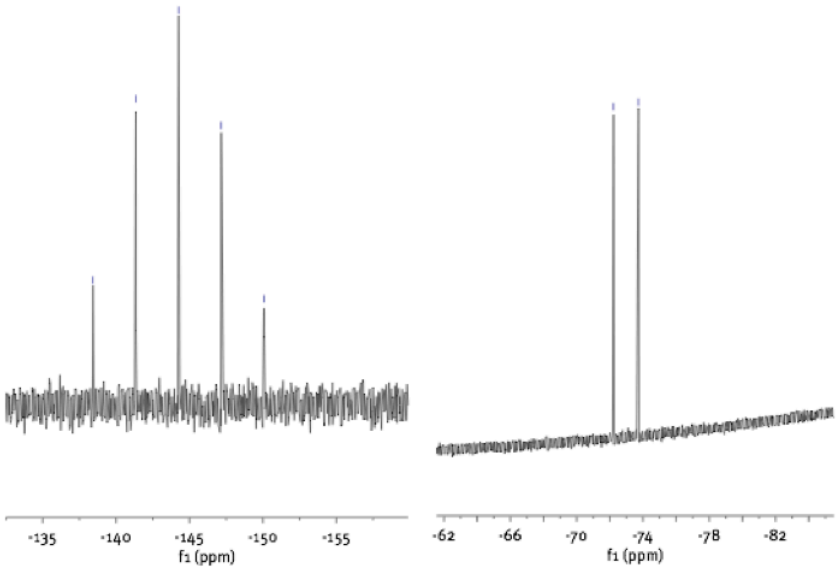


Figure S7. Partial ^{31}P NMR spectrum (CD_3CN, 243 MHz) of **2•PF$_6$** isolated from the bulk photolysis experiment (left) and ^{19}F NMR spectrum (CD_3CN, 471 MHz) of the same salt (right).

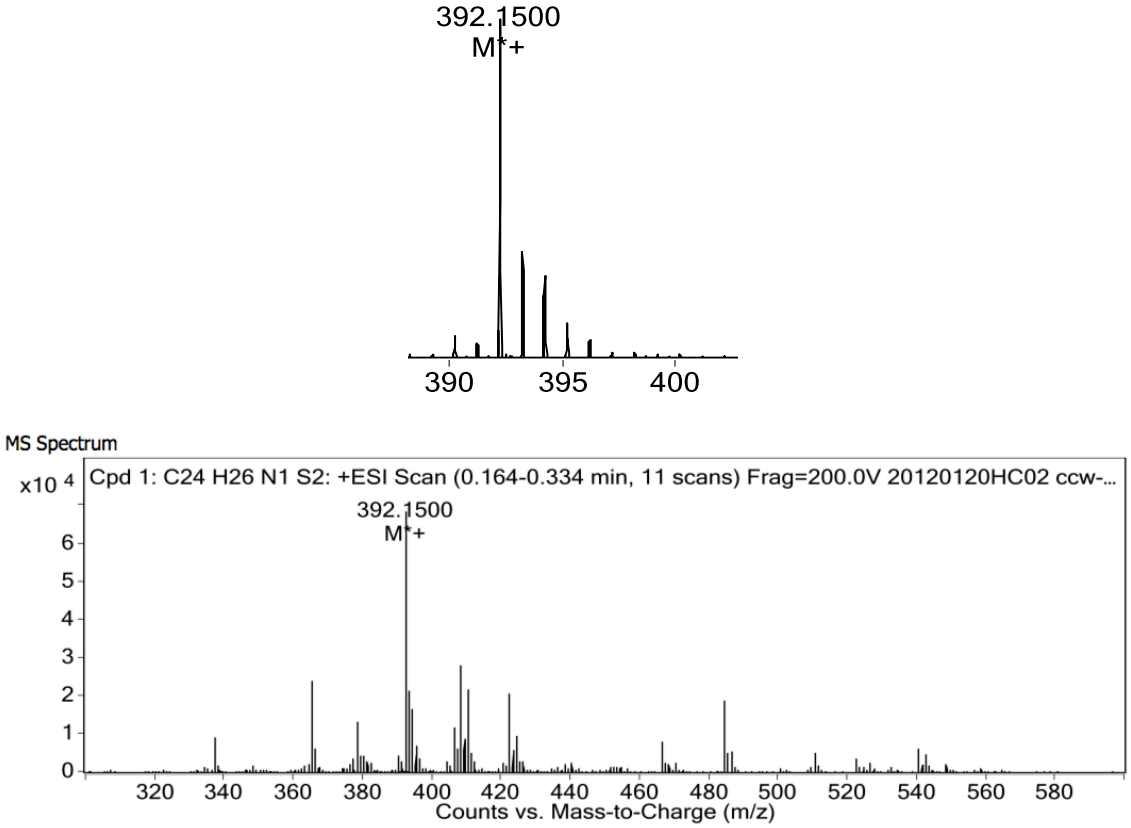


Figure S8. ESI-(+) HRMS analysis of **2•PF$_6$** isolated from the bulk photolysis experiment. The full analysis is shown at the bottom.