JOC The Journal of Organic Chemistry

Article

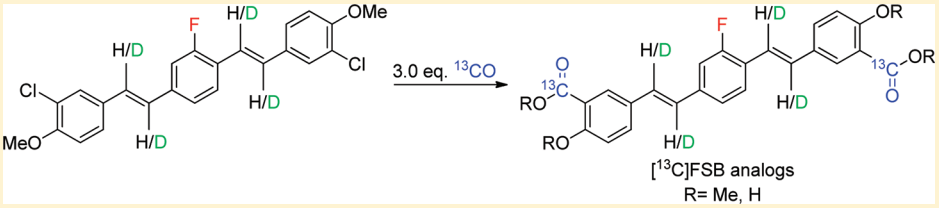
pubs.acs.org/joc

Isotope-Labeling of the Fibril Binding Compound FSB via a Pd-Catalyzed Double Alkoxycarbonylation

Mia N. Burhardt, Rolf Taaning, Niels Chr. Nielsen, and Troels Skrydstrup*

Center for Insoluble Protein Structures, Department of Chemistry and Interdisciplinary Nanoscience Center, Aarhus University, Langelandsgade 140, 8000 Aarhus C, Denmark

Supporting Information



ABSTRACT: We have synthesized two isotopically labeled variants of the β-amyloid binding compound FSB possessing ^{13}C-labels on the two terminal aryl carboxylic acid moieties. One of these was also fully deuterated on the olefinic spacers. The ^{13}C-isotope labeling was achieved applying a Pd-catalyzed methoxycarbonylation of the corresponding aryl chlorides with externally (ex situ) generated ^{13}C-labeled CO. Application of the Shirakawa–Hayashi protocol for the Pd-catalyzed reduction of a dialkyne intermediate using D_2O allowed for the selective deuterium labeling of the two *trans*-C,C double bonds of FSB.

INTRODUCTION

Alzheimer's disease (AD) is a neurodegenerative disorder characterized by the formation of β-amyloid fibrils (Aβ) and neurofibrillary tangles, with common symptoms such as memory impairment and disorientation.[1] AD is becoming a greater problem worldwide, with an estimate in 2006 of more than 24 million people, mostly elderly, suffering from this disease.[2] Currently, there is no known cure, and the only available treatment is temporary symptomatic relief of the cognitive decline.[3] In a patient suffering from AD, brain atrophy has usually been progressing for some time before the first AD symptoms are detectable. Since brain tissue cannot regenerate, it is crucial to develop effective techniques for early stage detection of AD leading to an early treatment of patients. Good tracers are also very important for the pharmaceutical industry in the evaluation of in vivo testing of new drug candidates in the search for a cure of this devastating disease.[4]

The styryl benzenes (ISB, BSB, FSB) were developed as ligands for Aβ recognition with the ability to cross the blood–brain barrier.[5] FSB ((*E*,*E*)-1-fluoro-2,5-bis-(3-hydroxycarbonyl-4-hydroxy)styrylbenzene) has affinity for both hallmarks of AD; Aβ and neurofibrillary tangles.[6] In 2005, Higuchi and co-workers demonstrated that ^{19}F-MRI in combination with the use of FSB could be applied for detecting Alzheimer plaques in mice models.[7] Whether FSB will be the optimal tracer for the diagnostics of Alzheimer plaques in brain tissue remains to be seen, but in order to design active and selective tracers for early stage AD detection, as well as active compounds for the dissolution of such pathological fibrils, it is important to obtain information about the molecular recognition events of binders such as FSB to β-amyloid fibrils. Solid-state NMR studies could be ideal for such a purpose, but it would require the introduction of NMR-active nuclei other than protons in different parts of FSB. ^{13}C-Labeling at specific sites of FSB would allow ^{13}C–^{13}C distance measurements between the core of FSB and labeled Aβ in solid-state NMR experiments. On the other hand, deuterium labeling of the FSB C,C-double bonds can possibly lead to information about the proximity of this region to the fibril via ^{2}H MAS experiments, whereby selective magnetization of the deuterium atoms can be transferred to the nondeuterated fibril.[8] Hence, we set out to prepare two isotopically labeled FSB compounds **1b** and **1c**, which would meet our needs for carrying out the above-described NMR experiments. In this paper, we demonstrate the successful synthesis of such isotopically labeled FSB derivatives using a new and simple carbonylation protocol developed in our group for the installment of ^{13}C-labels into bioactive compounds from externally generated ^{13}C-labeled carbon monoxide.[9] In combination with the Shirakawa–Hayashi protocol for the selective formation of deuterated *trans*-alkenes from alkynes, we also provide the synthesis of a hexa-isotopically labeled variant of FSB.

RESULTS AND DISCUSSION

FSB has previously been synthesized via three different routes (Figure 1), including procedures involving a Wittig–Horner reaction,[6a] a Pd-catalyzed double Mizoroki–Heck coupling,[10] and a high-yielding palladium-catalyzed double-Sonogashira coupling followed by a stereoselective Shirakawa–Hayashi reduction as the key steps for assembling the conjugated bis-stilbene system.[11] The latter two routes were developed in our

Received: April 24, 2012
Published: May 21, 2012

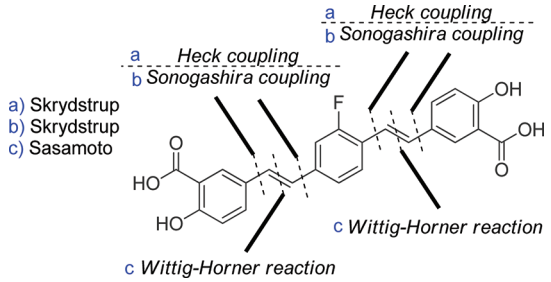


Figure 1. Previously reported routes to FSB.

group. In particular, the last protocol we felt would be suitable for the introduction of the various labels required for the fibril binding studies as the carbon backbone could be assembled in few synthetic steps and in high yields and subsequently allow for the introduction of four olefinic deuterium atoms applying the Shirakawa–Hayashi protocol for alkyne reduction.[12] In Scheme 1, we illustrate a retrosynthetic scheme for accessing FSB which is labeled with ^{13}C on the carboxylic acid moieties. The Sonogashira protocol (b in Figure 1) had to be modified to allow for a late-stage introduction of the ^{13}C-labeled carboxylic groups, thus aiming for an initial complete assembly of the dialkyne **3** which could then subsequently be reduced to the bis-stilbene **1** using the Shirakawa-Hayashi protocol (Scheme 1). Installment of the two labeled carboxylic acid groups was planned using a Pd-catalyzed alkoxycarbonylation combined with our carbonylation system using externally generated ^{13}C-labeled carbon monoxide. This method relies on the use of a closed two-chamber system (COware) whereby a slight excess of ^{13}CO generated externally from [^{13}C]-9-methyl-9*H*-fluorene-9-carbonyl chloride (^{13}C-COgen) is applied.[9a,13] In order to control the selectivity of the Sonogashira couplings, chlorides were chosen as the leaving groups for the introduction of the two carboxylic acids. Although the use of a Pd-catalyzed hydroxycarbonylation would provide the diacid, our previous experience with FSB revealed the compound to be cumbersome to purify becaue of its insolubility in common organic solvents and water. Hence, to facilitate purification of these FSB derivatives after the carbonylation step, we relied on the use of an analogous alkoxycarbonylation.

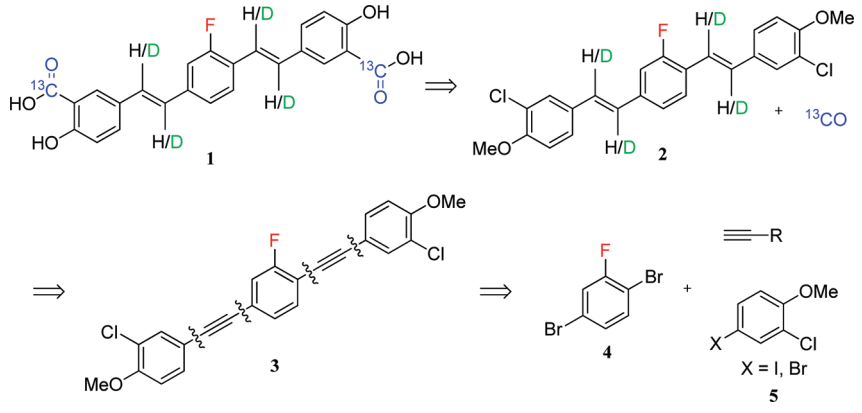
The 1,4-bis(phenylethynyl)benzene scaffold **3** was initially constructed in three steps from 1,4-dibromo-2-fluorobenzene (**4**) by first a Sonogashira coupling with TMS-acetylene followed by desilylation and then a double Sonogashira coupling with the iodide **5a** providing the diacetylene **3** in a good overall yield of 77%.[11] A more direct approach toward **3** would be to assemble the structure using the double one-pot Sonogashira coupling developed by Lee and co-workers.[14] Subjecting dibromobenzene **4** to 2 equiv of propiolic acid with the reported catalytic system $Pd(PPh_3)_2Cl_2$/dppb, DBU as a base in DMSO at 50 °C, followed by the addition of 2-chloro-4-iodoaniline (**5a**) at 80 °C only led to traces of **3**. Addition of 2-chloro-4-bromoaniline (**5b**) as the second substrate under the same conditions also proved futile. Lee has reported that the ligands dppb, dppf, and P^tBu_3 are almost equally effective for this reaction.[14] With this in mind, dppf and P^tBu_3 were used as ligands with otherwise identical reaction conditions. This was performed with both 2-chloro-4-iodoaniline (**5a**) and 2-chloro-4-bromoaniline (**5b**) as the second substrate, and gratifyingly, we found that the use of 2-chloro-4-iodoaniline (**5a**) and dppf as the ligand resulted in the isolation of **3** in a 43% yield (Scheme 2). This corresponds to an impressive 81% yield per C–C bond formation for the four generated C–C bonds.

Having constructed the carbon skeleton of FSB, the bis-styryl benzene **2** was obtained using the Shirakawa–Hayashi reduction with either H_2O (**2a**) or D_2O (**2b**) as the hydride source. The desired compounds were isolated in satisfying yields of 96% and 99%, respectively.

With the bis-styryl benzene precursors **2a** and **2b** in hand, we turned to the double Pd-catalyzed double alkoxycarbonylation. Our carbonylation conditions as previously reported for aryl bromides could not be directly applied to the aryl chlorides. In general, such compounds require higher reaction temperatures for carrying out such transformations. This required some optimization with our two-chamber system, since (a) we required a suitable solvent in the CO releasing chamber, which would not be volatile at the high reaction temperature (120 °C) used and thereby would not condense into the CO consuming chamber, and (b) the new high boiling solvent should not inhibit the decarbonylation reaction for CO production. Hence, a small screening of the CO-generating system applying deactivated chlorides at 120 °C was conducted, the results of which are shown in Table 1.

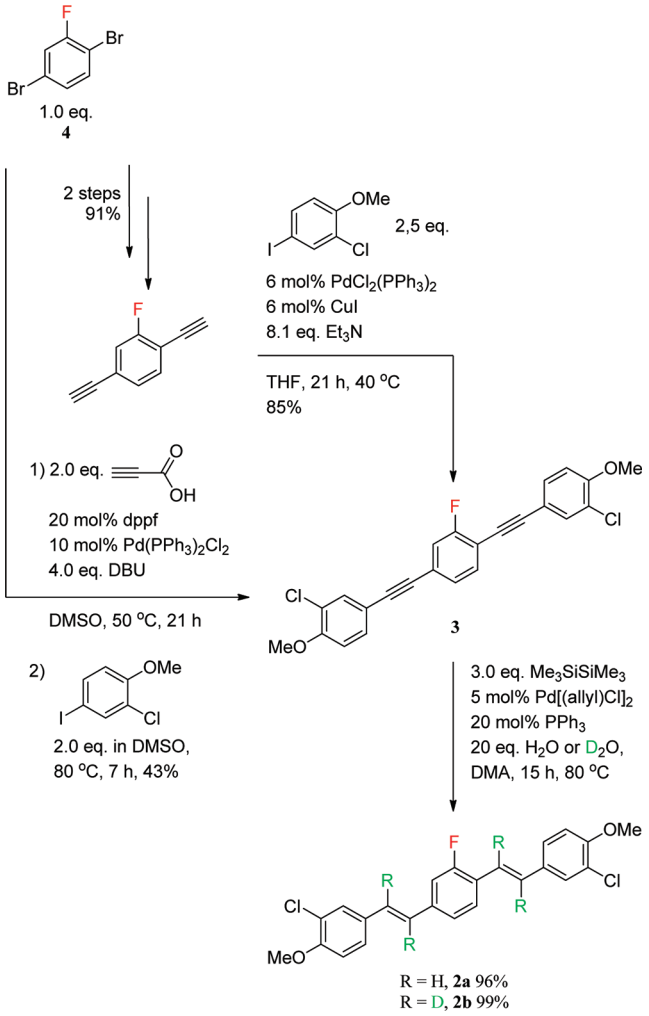
The alkoxycarbonylation of aryl chlorides has been widely studied.[15] The conditions chosen for the alkoxycarbonylation in chamber 1 were based on a mild protocol using CO at atmospheric pressure developed by Buchwald and co-workers.[15c] Three high-boiling solvents were tested in chamber 2 (entries 1–3). The use of paraffin wax and mesitylene as solvents in chamber 2 resulted in moderate conversions of the starting chloride in chamber 1 to the desired methyl ester **6**

Scheme 1. Retrosynthesis Analysis of an Isotopically Labeled FSB



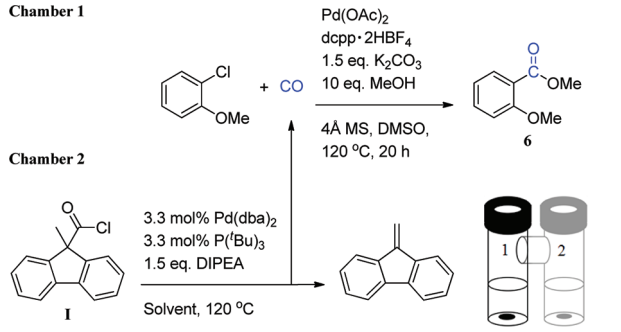
dx.doi.org/10.1021/jo300746x | J. Org. Chem. 2012, 77, 5357–5363

Scheme 2. Construction of the FSB Backbone



(52% and 45%, respectively) with isolated yields of 36 and 32%, respectively. However, when PEG 5000 was applied as the solvent in chamber 2, the carbonylation reaction afforded an 83% conversion of starting material and a 58% isolated yield of methyl ester **6**. In all cases, only 1.5 equiv of the CO precursor was used for these carbonylation reactions. To investigate this observed difference between the solvents examined, the rate of CO release was measured using a gas-volumetric apparatus.[16] The three solvents proved to have different effects on the rate of CO release at the desired temperature. The fastest CO release (85% within 2 min) was seen with PEG 5000 as the solvent (entry 3), whereas the CO-release in paraffin wax and mesitylene had only reached 65% and 58% completion, respectively, after 25 min (entries 1 and 2). These results in combination with the yields obtained for **6** suggest that the alkoxycarbonylations run best with a fast release of carbon monoxide from chamber 2. As a control experiment, 1.7 equiv of carbon monoxide was injected directly into a single reaction chamber using a gastight syringe prior to heating to 120 °C (entry 4). A 55% isolated yield of **6** was obtained, which was comparable to the results obtained when using PEG 5000 as the solvent for the CO release. Finally, to increase the coupling yield, we observed that by increasing the catalyst loading from 2 to 4 mol %, a 100% conversion was obtained affording the desired compound **6** in a 74% isolated yield (entry 5).

Table 1. Studies on the Methoxycarbonylation of *o*-Methoxychlorobenzene[a]



entry	solvent in chamber 2	NMR conv (%) (isolated yield, %)	CO release (%)	reaction time CO release (min)
1	Paraffin wax[b]	52 (36)	65	25[c]
2	Mesitylene[d]	45 (32)	58	25[c]
3	PEG 5000[e]	83 (58)	85	2
4[f]		70 (55)		
5[g]	PEG 5000[e]	100 (74)	85	2

[a]Reaction conditions: Chamber 1: 2-chloroanisole (0.50 mmol), MeOH (5.00 mmol), K_2CO_3 (0.75 mmol), dcpp·2HBF$_4$ (20 μmol), Pd(OAc)$_2$ (10 μmol), DMSO (0.5 mL), 4 Å MS (70 mg). Chamber 2: Pd(dba)$_2$ (33 μmol), P(^{t}Bu)$_3$ (33 μmol), DIPEA (1.13 mmol), 9-methyl-9H-fluorene-9-carbonyl chloride (0.75 mmol, 1.5 equiv). [b]0.50 g of Paraffin wax (mp 70−80 °C). [c]The CO release was not run to completion. [d]3 mL of mesitylene. [e]0.50 g of PEG 5000 monomethyl ether (PEG 5000). [f]1.7 equiv of CO gas was injected directly into the reaction vessel. [g]dcpp·2HBF$_4$ (40 μmol), Pd(OAc)$_2$ (20 μmol).

Applying these carbonylation conditions using 3 equiv of COgen to the bis-styryl benzene derivative **2a** afforded the tetra-*O*-methylated FSB derivative **7a**. Merely by changing the carbon monoxide precursor to the ^{13}C-COgen furnished the [^{13}C]-labeled intermediates **7b** and **7c** in good isolated yields (44% and 46%, respectively), considering that two carbon monoxide molecules are inserted into the dichlorides **2a** and **2b** (Scheme 3). The major byproduct observed in this reaction were determined to be the dehalogenated compound at both positions and the product resulting from only one carbonylation and a dehalogenation of the remaining chloride.

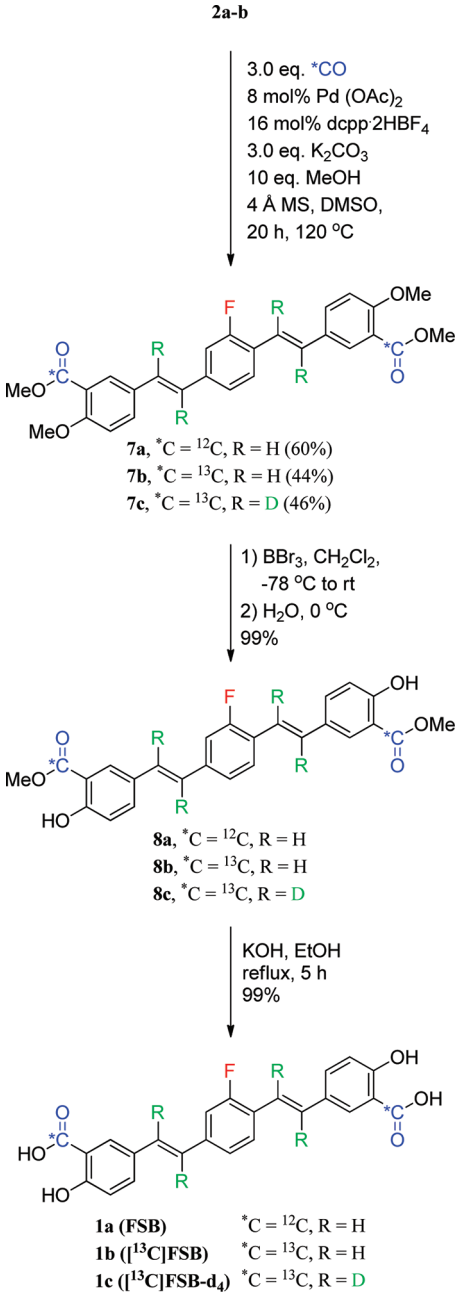
Subsequently, a sequential deprotection was performed with an initial selective demethylation of the two aryl methyl ethers using BBr_3 to afford **8a−c** in quantitative yields.[5] This was finally followed by basic hydrolysis of the methyl esters to obtain the fully deprotected FSB, [^{13}C]FSB, and [^{13}C]FSB-d_4 in quantitative yields from **7a−c**.

CONCLUSION

In summary, we have demonstrated a simple and useful protocol for the synthesis of the fibril binding compound FSB with ^{13}C- and ^{2}H-labeling. To achieve the ^{13}C-isotope-labeling, we applied our new carbonylation technique composed of a two-chamber system with a carbon monoxide releasing compound. This technique provides a useful method for performing simple and stoichiometric carbonylations, and in this work we have extended its use to aryl chlorides whereby the CO release could be effectively performed using a PEG solvent. Further work is ongoing to study the use of these isotopically labeled fibril-binding compounds in structural determination of their binding with β-amyloid using solid-state NMR. These results will be reported in due course.

dx.doi.org/10.1021/jo300746x | J. Org. Chem. 2012, 77, 5357−5363

Scheme 3. ^{13}C-Labeling of FSB



EXPERIMENTAL PROCEDURES

General Methods. Dry solvents were prepared according to standard literature procedures,[17] and all other chemicals were used as received from the suppliers unless mentioned otherwise. Starting materials were prepared according to literature procedures. Flash column chromatography was carried out on silica gel 60 (230−400 mesh). ^{1}H, ^{13}C, and ^{19}F NMR spectra were recorded at 400, 100 and 377 MHz, respectively. Chemical shifts are reported in ppm downfield to TMS (δ = 0) and referenced to the solvent residual peak.[18] HRMS was recorded on a LC TOF (ES). LRMS was recorded on a MALDI-TOF apparatus using DHBA (2,5-dihydrobenzoic acid) as the matrix.

4,4′-((2-Fluoro-1,4-phenylene)bis(ethyne-2,1-diyl))bis(2-chloro-1-methoxybenzene) (3). 1,4-Diethynyl-2-fluorobenzene (0.329 g, 2.28 mmol), 2-chloro-4-iodo-1-methoxybenzene (1.53 g, 5.70 mmol), CuI (0.026 g, 0.14 mmol), and $PdCl_2(PPh_3)_2$ (0.096 g, 0.14 mmol) were mixed and dissolved in THF (13 mL) in a glovebox under argon atmosphere followed by addition of Et_3N (2.6 mL, 19 mmol). The reaction mixture was removed from the glovebox and stirred overnight at 45 °C where it was diluted with Et_2O (50 mL) and CH_2Cl_2 (50 mL). The organic phase was washed with saturated NH_4Cl solution (3 × 100 mL), dried with Na_2SO_4, filtered, and concentrated in vacuo. The reaction was run in two parallel runs, and the crude reaction mixtures were combined during workup. The crude product was purified by flash chromatography on silica gel using pentane/CH_2Cl_2 (4:1) as eluent. This afforded the title compound **3** as a pale yellow solid (1.65 g, 3.89 mmol, 85%): mp 195.7−198.5 °C; ^{1}H NMR (400 MHz, $CDCl_3$) δ (ppm) 7.56 (dd, J = 4 Hz, J = 8 Hz, 1H), 7.45−7.38 (m, 3H), 7.26−7.20 (m, 3H), 6.89 (d, J = 8 Hz, 2H), 3.92 (s, 6H); ^{13}C NMR (100 MHz, $CDCl_3$) δ (ppm) 162.3 (d, J_{C-F} = 250 Hz), 155.9 (2C), 133.5 (2C), 133.3 (d, J_{C-F} = 1.8 Hz), 131.7, 131.6, 127.4 (d, J_{C-F} = 3.2 Hz), 125.0 (d, J_{C-F} = 9.4 Hz), 122.9 (2C), 118.5 (d, J_{C-F}= 22.2 Hz), 116.1, 115.9, 112.3, 112.2, 112.1, 95.0 (d, J_{C-F} = 3.0 Hz), 91.0, 87.9 (d, J_{C-F} = 3.3 Hz), 82.4, 56.4 (2C); ^{19}F NMR (377 MHz, $CDCl_3$) δ (ppm) −110.4 (t, J = 7.5 Hz); LRMS (MALDI) m/z calcd for $C_{24}H_{15}Cl_2FO_2$ [M + H$^+$] 425.1, found 425.5. Compound **3** did not provide a signal on the LC-TOF instrument and hence a HRMS of this compound was not possible.

4,4′-((2-Fluoro-1,4-phenylene)bis(ethyne-2,1-diyl))bis(2-chloro-1-methoxybenzene) (3). One-Pot Synthesis. 1,4-Dibromo-2-fluorobenzene (64 mg, 0.25 mmol), propiolic acid (30.8 μL, 0.500 mmol), dppf (28 mg, 0.050 mmol), and $PdCl_2(PPh_3)_2$ (18 mg, 0.025 mmol) were mixed and dissolved in DMSO (2.5 mL) in a glovebox under argon atmosphere followed by addition of DBU (149 μL, 1.00 mmol). The reaction mixture was removed from the glovebox and stirred overnight at 50 °C. 2-Chloro-4-iodo-1-methoxybenzene (134 mg, 0.500 mmol) was dissolved in DMSO (2 mL) under argon atmosphere and added to the reaction mixture. The temperature was raised to 80 °C and stirred for 7 h. The reaction mixture was diluted with CH_2Cl_2 (20 mL) and saturated NH_4Cl solution (20 mL) was added, followed by extraction of the water phase with CH_2Cl_2 (20 mL). The combined organic phase was then washed with H_2O (2 × 20 mL) and brine (1 × 20 mL), dried with Na_2SO_4, filtered, and concentrated in vacuo. The crude product was purified by flash chromatography on silica gel using pentane/CH_2Cl_2 (3:1) as eluent. This afforded the title compound **3** as a pale yellow solid (45 mg, 0.11 mmol, 43%).

***(E,E)*-1-Fluoro-2,5-bis(3-chloro-4-methoxy)styrylbenzene (2a).** 4,4′-((2-Fluoro-1,4-phenylene)bis(ethyne-2,1-diyl))bis(2-chloro-1-methoxybenzene) (**3**) (0.800 g, 1.88 mmol), hexamethyldisilane (1.2 mL, 5.6 mmol), H_2O (0.70 mL, 38 mmol), $[PdCl(\eta^3\text{-}C_3H_5)]_2$ (0.034 g, 0.094 mmol), and PPh_3 (0.099 g, 0.38 mmol) were mixed and dissolved in DMA (8 mL) in a glovebox under argon atmosphere. The reaction mixture was removed from the glovebox and stirred overnight at 80 °C where it was diluted with Et_2O (50 mL) and CH_2Cl_2 (50 mL). The organic phase was washed with H_2O (100 mL), the water phase was extracted with CH_2Cl_2, and then the organic phases were joined and washed with brine (100 mL). The organic phase was then dried with Na_2SO_4, filtered, and concentrated in vacuo. The crude product was purified by flash chromatography on silica gel using pentane/CH_2Cl_2 (1:1) as eluent. This afforded the title compound **2a** as a yellow solid (0.778 g, 1.81 mmol, 96%): mp 218.9−224.9 °C; ^{1}H NMR (400 MHz, $CDCl_3$) δ (ppm) 7.57 (dd, J = 8 Hz, J = 4 Hz, 2H), 7.53 (t, J = 8 Hz, 1H), 7.36 (ddd, J = 12 Hz, J = 12 Hz, J = 4 Hz, 2H), 7.25−6.89 (m, 8H), 3.93 (s, 6H); ^{1}H NMR (400 MHz, C_6D_6) δ (ppm) 7.52 (d, J = 2 Hz, 2H), 7.33 (m, 1H), 7.03 (m, 6H), 6.75 (d, J = 16 Hz, 1H), 6.65 (d, J = 16 Hz, 1H), 6.39 (d, J = 16 Hz, 1H), 6.37 (d, J = 16 Hz, 1H), 3.25 (s, 1H), 3.22 (s, 1H); ^{19}F NMR (377 MHz, $CDCl_3$) δ (ppm) −118.5 (dd, J = 11.3 Hz, J = 7.5 Hz); LRMS (MALDI) m/z calcd for $C_{24}H_{19}Cl_2FO_2$ [M + H$^+$] 429.1, found 429.5. ^{13}C NMR could not be aquired due to solubility issues in all common deuterated solvents. Full assignment of the ^{1}H NMR in C_6D_6 was not attained due to poor solubility. Compound **2a** did not provide a signal on the LC-TOF instrument, and hence, an HRMS of this compound was not possible.

***(E,E)*-1-Fluoro-2,5-bis(3-chloro-4-methoxy)-1,1′,2,2′-tetradeuterostyrylbenzene (2b).** 4,4′-((2-Fluoro-1,4-phenylene)bis(ethyne-2,1-diyl))bis(2-chloro-1-methoxybenzene) (**3**) (0.400 g, 0.941 mmol), hexamethyldisilane (0.58 mL, 2.8 mmol), D_2O (0.34

dx.doi.org/10.1021/jo300746x | J. Org. Chem. 2012, 77, 5357−5363

mL, 19 mmol), $[PdCl(\eta^3\text{-}C_3H_5)]_2$ (0.017 g, 0.047 mmol), and PPh_3 (0.049 g, 0.19 mmol) were mixed and dissolved in DMA (4 mL) in a glovebox under argon atmosphere. The reaction mixture was removed from the glovebox and stirred overnight at 80 °C, and then it was diluted with Et_2O (25 mL) and CH_2Cl_2 (25 mL). The organic phase was washed with H_2O (50 mL), the water phase was extracted with CH_2Cl_2, and then the organic phases were joined and washed with brine (50 mL). The organic phase was then dried with Na_2SO_4, filtered, and concentrated in vacuo. The crude product was purified by flash chromatography on silica gel using pentane/CH_2Cl_2/EtOAc (30:10:1) as eluent. This afforded the title compound **2b** as a yellow solid (0.40 g, 0.93 mmol, 99%): mp 216.8−223.6 °C; ^{1}H NMR (400 MHz, $CDCl_3$) δ (ppm) 7.57 (dd, J = 8 Hz, J = 4 Hz, 2H), 7.53 (t, J = 8 Hz, 1H), 7.36 (ddd, J = 12 Hz, J = 12 Hz, J = 4 Hz, 2H), 7.25−7.17 (m, 2H), 6.92 (d, J = 8 Hz, 2H), 3.93 (s, 6H); ^{19}F NMR (377 MHz, $CDCl_3$) δ (ppm) −118.6 (dd, J = 15 Hz, J = 7.5 Hz); LRMS (MALDI) m/z calcd for $C_{24}H_{15}D_4Cl_2FO_2$ [M + H$^+$] 433.1, found 433.6. Compound **2b** did not provide a signal on the LC-TOF instrument, and hence, a HRMS of this compound was not possible.

General Procedure for the Alkoxycarbonylation of 2a and 2b. The alkoxycarbonylation of **2a** and **2b** was performed in a two-chamber reaction vessel with a total volume of 20 mL using either 9-methylfluorene-9-carbonyl chloride or 9-methylfluorene-9-[^{13}C]-carbonyl chloride as the source of carbon monoxide. The two chambers were loaded in an argon filled glovebox. Chamber 1 was loaded with the substrates and reactants needed for the alkoxycarbonylation, and chamber 2 was loaded with 9-methylfluorene-9-carbonyl chloride or 9-methylfluorene-9-[^{13}C]-carbonyl chloride, $Pd(dba)_2$ (3.3 mol %), P(t-Bu)$_3$ (3.3 mol %), PEG 5000 monomethyl ether (0.5 g), and DIPEA. The chambers were sealed with a screw cap, 2 mm stabilizing PTFE disk, and a 2 mm thick PTFE-lined silicone disk before taking it out of the glovebox and heating at the appropriate temperature.

(*E,E*)-1-Fluoro-2,5-bis(3-methoxycarbonyl-4-methoxy)-styrylbenzene (7a). Chamber 1: (*E,E*)-1-Fluoro-2,5-bis(3-chloro-4-methoxy)styrylbenzene (**2a**) (107 mg, 0.250 mmol), MeOH (203 μL, 5.00 mmol), K_2CO_3 (104 mg, 0.750 mmol), dcpp·2HBF$_4$ (25 mg, 0.040 mmol), $Pd(OAc)_2$ (5.0 mg, 0.020 mmol), and pulverized 4 Å molecular sieves (140 mg) were mixed in DMSO (1 mL). Chamber 2: $Pd(dba)_2$ (14 mg, 0.025 mmol), P(t-Bu)$_3$ (5.0 mg, 0.025 mmol), 9-methylfluorene-9-carbonyl chloride (182 mg, 0.750 mmol), PEG 5000 monomethyl ether (0.5 g), and DIPEA (196 μL, 1.13 mmol). The reaction system was removed from the glovebox and stirred for 20 h at 120 °C, and then it was diluted with Et_2O (20 mL) and CH_2Cl_2 (20 mL) and filtered through Celite. The filtrate was washed with H_2O (2 × 20 mL) and brine (1 × 20 mL), dried with Na_2SO_4, filtered, and concentrated in vacuo. The crude product was purified twice by flash chromatography on silica gel using first CH_2Cl_2 and then CH_2Cl_2/pentane/EtOAc (5:5:1) as eluents. This afforded the title compound **7a** as a yellow solid (75 mg, 0.16 mmol, 63%): mp 179.0−182.2 °C; ^{1}H NMR (400 MHz, $CDCl_3$) δ (ppm) 7.96 (dd, J = 4.0 Hz, J = 2.4 Hz, 2H), 7.59 (ddd, J = 13.2 Hz, J = 8.8 Hz, J = 2.4 Hz, 2H), 7.52 (t, J = 8 Hz, 1H), 7.23−6.91 (m, 8H), 3.92 (s, 6H), 3.92 (s, 6H); ^{13}C NMR (100 MHz, $CDCl_3$) δ (ppm) 166.72, 166.70, 160.8 (d, J_{C-F} =249 Hz), 159.04, 159.01, 138.5 (d, J_{C-F} = 8.3 Hz), 131.74, 131.65, 130.13, 130.05, 129.9, 129.5, 129.3 (d, J_{C-F} = 4.9 Hz), 128.4 (2C), 127.3 (d, J_{C-F} = 4.3 Hz), 126.5 (d, J_{C-F} = 2.3 Hz), 124.4 (d, J_{C-F} = 12.6 Hz), 122.7 (d, J_{C-F} = 2.8 Hz), 120.5 (d, J_{C-F} = 3.2 Hz), 120.0 (d, J_{C-F} = 3.1 Hz), 113.3 (d, J_{C-F} = 23.2 Hz), 112.6 (2C), 56.4 (2C), 52.37, 52.36; ^{19}F NMR (377 MHz, $CDCl_3$) δ (ppm) −118.6 (dd, J = 12.1 Hz, J = 7.9 Hz); HRMS (ESI) m/z calcd for $C_{28}H_{25}FO_6$ [M + Na$^+$] 499.1533, found 499.1540.

(*E,E*)-1-Fluoro-2,5-bis(3-methoxy[^{13}C]carbonyl-4-methoxy)-styrylbenzene (7b). Chamber 1: (*E,E*)-1-Fluoro-2,5-bis(3-chloro-4-methoxy)-styrylbenzene (**2a**) (107 mg, 0.250 mmol), MeOH (203 μL, 5.00 mmol), K_2CO_3 (104 mg, 0.750 mmol), dcpp·2HBF$_4$ (25 mg, 0.040 mmol), $Pd(OAc)_2$ (5.0 mg, 0.020 mmol), and pulverized 4 Å molecular sieves (140 mg) were mixed in DMSO (1 mL). Chamber 2: $Pd(dba)_2$ (14 mg, 0.025 mmol), P(t-Bu)$_3$ (5.0 mg, 0.025 mmol), 9-methylfluorene-9-[^{13}C]-carbonyl chloride (183 mg, 0.750 mmol), PEG 5000 monomethyl ether (0.5 g), and DIPEA (196 μL, 1.13 mmol). The reaction system was removed from the glovebox and stirred for 20 h at 120 °C, and then it was diluted with Et_2O (20 mL) and CH_2Cl_2 (20 mL) and filtered through Celite. The filtrate was washed with H_2O (2 × 20 mL) and brine (1 × 20 mL), dried with Na_2SO_4, filtered, and concentrated in vacuo. The crude product was purified twice by flash chromatography on silica gel using first CH_2Cl_2 and then CH_2Cl_2/pentane/EtOAc (5:5:1) as eluents. This afforded the title compound **7b** as a yellow solid (54 mg, 0.11 mmol, 45%): mp 179.5−183.2 °C; ^{1}H NMR (400 MHz, $CDCl_3$) δ (ppm) 7.98−7.96 (m, 2H), 7.63 (ddd, J = 12.8 Hz, J = 8.8 Hz, J = 2.4 Hz, 2H), 7.54 (t, J = 8.0 Hz, 1H), 7.24−6.94 (m, 8H), 3.93 (s, 6H), 3.92 (s, 6H); ^{13}C NMR (100 MHz, $CDCl_3$) δ (ppm) 166.72 (^{13}C), 166.70 (^{13}C), 160.8 (d, J_{C-F} =248.8 Hz), 159.05, 159.02, 138.5 (d, J_{C-F} = 8.3 Hz), 131.74, 131.66, 130.15, 130.07, 129.9 (d, $J_{C-^{13}C}$ = 5.0 Hz), 129.5 (d, $J_{C-^{13}C}$ = 4.0 Hz), 129.4 (d, J_{C-F} = 5.0 Hz), 128.4 (2C), 127.3 (d, J_{C-F} = 4.0 Hz), 126.5 (d, J_{C-F} = 2.0 Hz), 124.4 (d, J_{C-F} = 12.6 Hz), 122.7 (d, J_{C-F} = 2.8 Hz), 120.9 (d, J_{C-F} = 3.2 Hz), 120.0 (d, J_{C-F} = 3.0 Hz), 113.3 (d, J_{C-F} = 23.2 Hz), 112.6 (2C), 56.4 (2C), 52.4 (2C); ^{19}F NMR (377 MHz, $CDCl_3$) δ (ppm) −118.6 (dd, J = 11.3 Hz, J = 7.5 Hz); HRMS (ESI) m/z calcd for $C_{26}{}^{13}C_2H_{25}FO_6$ [M + Na$^+$] 501.1600, found 501.1588.

(*E,E*)-1-Fluoro-2,5-bis(3-methoxy[^{13}C]carbonyl-4-methoxy)-1,1′,2,2′-tetradeuterostyrylbenzene (7c). Chamber 1: (*E,E*)-1-Fluoro-2,5-bis(3-chloro-4-methoxy)-1,1′,2,2′-tetradeutero-styrylbenzene (**2b**) (108 mg, 0.250 mmol), MeOH (203 μL, 5.00 mmol), K_2CO_3 (104 mg, 0.750 mmol), dcpp·2HBF$_4$ (25 mg, 0.040 mmol), $Pd(OAc)_2$ (5.0 mg, 0.020 mmol), pulverized 4 Å molecular sieves (140 mg) were mixed in DMSO (1 mL). Chamber 2: $Pd(dba)_2$ (14 mg, 0.025 mmol), P(t-Bu)$_3$ (5.0 mg, 0.025 mmol), 9-methylfluorene-9-[^{13}C]-carbonyl chloride (183 mg, 0.750 mmol), PEG 5000 monomethyl ether (0.5 g) and DIPEA (196 μL, 1.13 mmol). The reaction system was removed from the glovebox and stirred for 20 h at 120 °C where after it was diluted with Et_2O (20 mL) and CH_2Cl_2 (20 mL) and filtered through Celite. The filtrate was washed with H_2O (2 × 20 mL) and brine (1 × 20 mL), dried with Na_2SO_4, filtered, and concentrated in vacuo. The crude product was purified twice by flash chromatography on silica gel using first CH_2Cl_2 and then CH_2Cl_2/Pentane/EtOAc (5:5:1) as eluents. This afforded the title compound **7c** as a yellow solid (69 mg, 0.14 mmol, 46%): mp 177.8−180.7 °C; ^{1}H NMR (400 MHz, $CDCl_3$) δ (ppm) 7.97−7.96 (m, 2H), 7.60 (ddd, J = 12.8 Hz, J = 8.4 Hz, J = 2.0 Hz, 2H), 7.53 (t, J = 8.0 Hz, 1H), 7.23 (dd, J = 8.0 Hz, J = 1.2 Hz, 1H), 7.18 (dd, J = 12.0 Hz, J = 1.2 Hz, 1H), 6.97 (dd, J = 8.8 Hz, J = 1.6 Hz, 2H), 3.93 (s, 6H), 3.92 (s, 6H); ^{13}C NMR (100 MHz, $CDCl_3$) δ (ppm) 166.49 (^{13}C), 166.46 (^{13}C), 160.6 (d, J_{C-F} =249 Hz), 158.79, 158.76, 138.2 (d, J_{C-F} = 8.1 Hz), 131.5, 131.4, 129.9, 129.8, 129.6 (d, $J_{C-^{13}C}$ = 5.0 Hz), 129.2 (d, $J_{C-^{13}C}$ = 4.0 Hz), 127.0 (d, J_{C-F} = 4.0 Hz), 124.1 (d, J_{C-F} = 13.1 Hz), 122.4 (d, J_{C-F} = 3.0 Hz), 120.6 (d, J_{C-F} = 3.0 Hz), 119.9 (d, J_{C-F} = 3.0 Hz), 113.1 (d, J_{C-F} = 22.2 Hz), 112.3 (2C), 56.2 (2C), 52.1 (2C); ^{19}F NMR (377 MHz, $CDCl_3$) δ (ppm) −118.5 (dd, J = 12.4 Hz, J = 8.3 Hz); HRMS (ESI) m/z calcd for $C_{26}{}^{13}C_2H_{21}D_4FO_6$ [M + Na$^+$] 505.1851, found 505.1850. The C−D carbon signals are so small due to the deuterium coupling that they disappear from the ^{13}C NMR spectrum.

(*E,E*)-1-Fluoro-2,5-bis(3-methoxycarbonyl-4-hydroxy)-styryl-benzene (8a). (*E,E*)-1-Fluoro-2,5-bis(3-methoxycarbonyl-4-methoxy)styrylbenzene (**7a**) (47 mg, 0.099 mmol) was dissolved in dry CH_2Cl_2 (15 mL) under argon atmosphere. The solution was cooled to −78 °C, and BBr_3 was added dropwise (1 M in hexane, 0.75 mL, 0.75 mmol). The mixture was subsequently allowed to warm to rt and cooled to 0 °C, at which point the reaction was quenched with H_2O. The mixture was extracted with CH_2Cl_2 (2 × 15 mL), and the organic phase washed with brine (2 × 15 mL), dried with Na_2SO_4, filtered, and concentrated in vacuo. This afforded the title compound **8a** as a yellow solid (42 mg, 0.094 mmol, 95%): mp 223 °C dec; ^{1}H NMR (400 MHz, $CDCl_3$) δ (ppm) 10.81 (s, 2H), 7.98 (dd, J = 7.6 Hz, J = 2.4 Hz, 2H), 7.67 (ddd, J = 10.8 Hz, J = 9.2 Hz, J = 2.4 Hz, 2H), 7.55 (t, J = 8.0 Hz, 1H), 7.24−6.91 (m, 8H), 4.00 (s, 6H); ^{13}C NMR (100 MHz, $CDCl_3$) δ (ppm) 170.33, 170.28, 161.43, 161.40, 160.6 (d, J_{C-F} =249 Hz), 138.3 (d, J_{C-F} = 8.3 Hz), 133.4, 133.4, 129.4 (d, J_{C-F} = 5.2 Hz), 128.9, 128.5, 128.4, 128.20, 128.18, 127.0 (d, J_{C-F} = 4.3 Hz),

dx.doi.org/10.1021/jo300746x | J. Org. Chem. 2012, 77, 5357−5363

126.0 (d, J_{C-F} = 2.5 Hz), 124.2 (d, J_{C-F} = 12.7 Hz), 122.4 (d, J_{C-F} = 2.7 Hz), 119.6 (d, J_{C-F} = 3.0 Hz), 118.14, 118.09, 113.0 (d, J_{C-F} = 23.0 Hz), 112.54, 112.51, 52.32, 52.30; ^{19}F NMR (377 MHz, $CDCl_3$) δ (ppm) −118.4 (dd, J = 12.1 Hz, J = 7.9 Hz); LRMS (MALDI) m/z calcd for $C_{26}H_{21}FO_6$ [M + H$^+$] 449.1, found 449.4.

(*E*,*E*)-1-Fluoro-2,5-bis(3-methoxy[^{13}C]carbonyl-4-hydroxy)-styrylbenzene (8b). Same procedure as described for **8a**. (*E*,*E*)-1-Fluoro-2,5-bis(3-methoxy[^{13}C]carbonyl-4-methoxy)styrylbenzene (**7b**) (54 mg, 0.11 mmol) and BBr_3 (1 M in hexane, 0.87 mL, 0.87 mmol) were dissolved in dry CH_2Cl_2 (17 mL). This afforded the title compound **8b** as a yellow solid (52 mg, 0.12 mmol, quantitative yield): mp 236 °C dec; ^{1}H NMR (400 MHz, $CDCl_3$) δ (ppm) 10.81 (s, 2H), 7.98 (ddd, J = 6.8 Hz, J_{C-H} = 4.4 Hz, J = 2.0 Hz, 2H), 7.67 (ddd, J = 10.8 Hz, J = 8.8 Hz, J = 2.4 Hz, 2H), 7.55 (t, J = 8.0 Hz, 1H), 7.24−6.91 (m, 8H), 4.00 (d, J_{C-H}= 4.0 Hz, 6H); ^{13}C NMR (100 MHz, $CDCl_3$) δ (ppm) 170.4 (^{13}C), 170.3 (^{13}C); ^{19}F NMR (377 MHz, $CDCl_3$) δ (ppm) −118.4 (dd, J = 11.3 Hz, J = 7.5 Hz); LRMS (MALDI) m/z calcd for $C_{24}{}^{13}C_2H_{21}FO_6$ [M + H$^+$] 451.1, found 451.8. Compound **8b** did not provide a signal on the LC-TOF instrument, and hence, a HRMS of this compound was not possible.

(*E*,*E*)-1-Fluoro-2,5-bis(3-methoxy[^{13}C]carbonyl-4-hydroxy)-1,1′,2,2′-tetradeuterostyrylbenzene (8c). Same procedure as described for **8a**. (*E*,*E*)-1-Fluoro-2,5-bis(3-methoxy[^{13}C]carbonyl-4-methoxy)-1,1′,2,2′-tetradeuterostyrylbenzene (**7c**) (55 mg, 0.12 mmol) and BBr_3 (1 M in hexane, 0.88 mL, 0.88 mmol) were dissolved in dry CH_2Cl_2 (17 mL). This afforded the title **8c** compound as a yellow solid (54 mg, 0.12 mmol, quantitative yield): mp 236 °C dec; ^{1}H NMR (400 MHz, $CDCl_3$) δ (ppm) 10.81 (s, 2H), 7.98 (ddd, J = 6.8 Hz, J_{C-H} = 4.4 Hz, J = 2.4 Hz, 2H), 7.67 (ddd, J = 10.4 Hz, J = 8.8 Hz, J = 2.0 Hz, 2H), 7.55 (t, J = 8.0 Hz, 1H), 7.24−7.18 (m, 2H), 7.00 (dd, J = 8.4 Hz, J = 1.2 Hz, 2H), 4.00 (d, J_{C-H}= 3.6 Hz, 6H); ^{13}C NMR (100 MHz, $CDCl_3$) δ (ppm) 170.4 (^{13}C), 170.3 (^{13}C); ^{19}F NMR (377 MHz, $CDCl_3$) δ (ppm) −118.4 (dd, J = 12.4 Hz, J = 7.9 Hz); LRMS (MALDI) m/z calcd for $C_{24}{}^{13}C_2H_{17}D_4FO_6$ [M + H$^+$] 455.5, found 455.2. Compound **8c** did not provide a signal on the LC-TOF instrument, and hence, a HRMS of this compound was not possible.

(*E*,*E*)-1-Fluoro-2,5-bis-(3-hydroxycarbonyl-4-hydroxy)-styrylbenzene (FSB) (1a). KOH (s) (0.10 g, 1.9 mmol) was added to a suspension of (*E*,*E*)-1-fluoro-2,5-bis(3-methoxycarbonyl-4-hydroxy)-styrylbenzene (**8a**) (39 mg, 0.087 mmol) in ethanol (96%, 5 mL). The mixture was refluxed for 5 h and allowed to cool to rt before the solvent was removed by evaporation. HCl (1 M) was added, and the slurry was transferred to a falcon tube. After shaking, the product was spun down and the supernatant removed. This was done twice. The product was then washed twice with H_2O and twice with pentane using the same procedure. Pentane traces was removed by evaporation before the product was left on the freeze-dryer overnight. This afforded the title compound **1a** as a yellow solid (33 mg, 0.078 mmol, 90%): mp 294 °C dec; ^{1}H NMR (400 MHz, $CDCl_3$) δ (ppm) 7.99 (dd, J = 8.0 Hz, J = 2.4 Hz, 2H), 7.84 (ddd, J = 15.6 Hz, J = 8.8 Hz, J = 2.0 Hz, 2H), 7.77 (t, J = 8.4 Hz, 1H),7.47 (d, J = 12.8 Hz, 1H), 7.44 (d, J = 8.4 Hz, 1H), 7.35 (dd, J = 16.4 Hz, J = 2.8 Hz, 2H), 7.15 (dd, J = 16.8 Hz, J = 2.4 Hz, 2H), 7.00 (dd, J = 8.8 Hz, J = 2.4 Hz, 2H); ^{19}F NMR (377 MHz, $CDCl_3$) δ (ppm) −118.8 (dd, J = 12.8 Hz, J = 8.3 Hz); LRMS (MALDI) m/z calcd for $C_{24}H_{17}FO_6$ [M + H$^+$] 421.1, found 421.3. The spectral data is consistent with previously reported data for this compound.[10]

(*E*,*E*)-1-Fluoro-2,5-bis-(3-hydroxy[^{13}C]carbonyl-4-hydroxy)-styrylbenzene ([^{13}C] FSB) (1b). Same procedure as described for **1a**. KOH (s) (0.14 g, 2.5 mmol) was added to a suspension of (*E*,*E*)-1-fluoro-2,5-bis(3-methoxy[^{13}C]carbonyl-4-hydroxy)styrylbenzene (**8b**) (53 mg, 0.12 mmol) in ethanol (96%, 7 mL). This afforded the title compound **1b** as a brown solid (45 mg, 0.11 mmol, 90%): mp 298 °C dec; ^{1}H NMR (400 MHz, $CDCl_3$) δ (ppm) 7.99 (ddd, J = 7.6 Hz, J_{C-H} = 4.4 Hz, J = 2.0 Hz, 2H), 7.82 (ddd, J = 14.4 Hz, J = 8.8 Hz, J = 2.0 Hz, 2H), 7.76 (t, J = 8.0 Hz, 1H),7.47 (d, J = 12.8 Hz, 1H), 7.43 (d, J = 8.4 Hz, 1H), 7.34 (dd, J = 16.0 Hz, J = 2.8 Hz, 2H), 7.13 (dd, J = 16.4 Hz, J = 2.4 Hz, 2H), 6.98 (d, J = 8.8 Hz, 2H); ^{13}C NMR (100 MHz, $CDCl_3$) δ (ppm) 172.1 (^{13}C), 172.0 (^{13}C); ^{19}F NMR (377 MHz, $CDCl_3$) δ (ppm) −118.8 (dd, J = 12.4 Hz, J = 8.3 Hz); LRMS (MALDI) m/z calcd for $C_{22}{}^{13}C_2H_{17}FO_6$ [M + H$^+$] 423.1, found 423.1. Compound **1b** did not provide a signal on the LC-TOF instrument, and hence, a HRMS of this compound was not possible.

(*E*,*E*)-1-Fluoro-2,5-bis-(3-hydroxy[^{13}C]carbonyl-4-hydroxy)-1,1′,2,2′-tetradeuterostyrylbenzene ([^{13}C] FSB-d$_4$) (1c). Same procedure as described for **1a**. KOH (s) (54 mg, 0.96 mmol) was added to a suspension of (*E*,*E*)-1-fluoro-2,5-bis(3-methoxy[^{13}C]-carbonyl-4-hydroxy)-1,1′,2,2′-tetradeuterostyrylbenzene (**8c**) (20 mg, 0.044 mmol) in ethanol (96%, 3 mL). This afforded the title compound **1c** as a yellow solid (21 mg, 0,048 mmol, quantitative yield): mp 294 °C dec; ^{1}H NMR (400 MHz, $CDCl_3$) δ (ppm) 7.98 (ddd, J = 8.4 Hz, J_{C-H} = 4.4 Hz, J = 2.4 Hz, 2H), 7.77 (m, 3H), 7.46 (dd, J = 12.8 Hz, J = 1.2 Hz, 1H), 7.43 (dd, J = 8.0 Hz, J = 1.6 Hz, 1H), 6.94 (ddd, J = 8.8 Hz, J = 3.2 Hz, J = 1.6 Hz, 2H); ^{13}C NMR (100 MHz, $CDCl_3$) δ (ppm) 172.1 (^{13}C), 172.0 (^{13}C); ^{19}F NMR (377 MHz, $CDCl_3$) δ (ppm) −118.9 (dd, J = 12.1 Hz, J = 8.3 Hz); LRMS (MALDI) m/z calcd for $C_{22}{}^{13}C_2H_{13}D_4FO_6$ [M + H$^+$] 427.1, found 427.5. Compound **1c** did not provide a signal on the LC-TOF instrument, and hence, a HRMS of this compound was not possible.

General Procedure for the Methoxycarbonylations of *o*-Methoxychlorobenzene (6). Chamber 1: 2-chloroanisole (64 μL, 0.50 mmol), MeOH (203 μL, 5.00 mmol), K_2CO_3 (104 mg, 0.750 mmol), dcpp·2HBF$_4$, Pd(OAc)$_2$, and pulverized 4 Å molecular sieves (70 mg) were mixed in DMSO (1 mL). Chamber 2: Pd(dba)$_2$ (14 mg, 0.025 mmol), P(t-Bu)$_3$ (5.0 mg, 0.025 mmol), 9-methylfluorene-9-carbonyl chloride (182 mg, 0.750 mmol), solvent (0.5 g for the solid solvents or 3 mL), and DIPEA (196 μL, 1.13 mmol). The reaction system was removed from the glovebox and stirred for 20 h at 120 °C, and then it was diluted with EtOAc (20 mL) and filtered through Celite. The filtrate was washed with H_2O (2 × 20 mL) and brine (1 × 20 mL), dried with Na_2SO_4, filtered, and concentrated in vacuo. The crude product was purified by flash chromatography on silica gel using CH_2Cl_2 as eluent. This afforded the title compound **6** as a colorless oil: ^{1}H NMR (400 MHz, $CDCl_3$) δ (ppm) 7.79 (dd, J = 8.0 Hz, J = 2.0 Hz, 1H), 7.46 (ddd, J = 9.2 Hz, J = 7.6 Hz, J = 2.0 Hz, 1H), 6.99−6.95 (m, 2H), 3.89 (s, 3H), 3.88 (s, 3H); HRMS (ESI) m/z calcd for $C_9H_{10}O_3$ [M + Na$^+$] 189.0528, found 189.0527. The spectral data are consistent with previously reported data for this compound.[19]

■ ASSOCIATED CONTENT

Ⓢ Supporting Information

Experimental details and copies of ^{1}H NMR, ^{13}C NMR, and ^{19}F NMR spectra for all of the coupling products. This material is available free of charge via the Internet at http://pubs.acs.org

■ AUTHOR INFORMATION

Corresponding Author

*E-mail: ts@chem.au.dk.

Notes

The authors declare no competing financial interest.

■ ACKNOWLEDGMENTS

We are deeply appreciative of generous financial support from the Danish National Research Foundation, the Danish Natural Science Research Council, the Carlsberg Foundation, the Lundbeck Foundation, iNANO, and Aarhus University.

■ REFERENCES

(1) Jakob-Roetne, R.; Jacobsen, H. *Angew. Chem., Int. Ed.* **2009**, *48*, 2.
(2) Jellinger, K. A. *J. Neural. Transm.* **2006**, *113*, 1603.
(3) Chopra, K.; Misra, S.; Kuhad, A. *Expert Opin. Pharmacother.* **2011**, *12*, 335.
(4) Halford, B. *Chem. Eng. News* **2008**, *86*, 13.
(5) (a) Zhuang, Z.-P.; Kung, M.-P.; Hou, C.; Skovronsky, D. M.; Gur, T. L.; Plössl, K.; Trojanowski, J. Q.; Lee, V. M.-Y.; Kung, H. F. *J. Med. Chem.* **2001**, *44*, 1905. (b) Lee, C.-W.; Zhuang, Z.-P.; Kung, M.-

P.; Plössl, K.; Skovronsky, D.; Gur, T.; Hou, C.; Trojanowski, J. Q.; Lee, M.-Y.; Kung, H. F *J. Med. Chem.* **2001**, *44*, 2270.
(6) (a) Sato, K.; Higuchi, M.; Iwata, N.; Saido, T. C.; Sasamoto, K. *Eur. J. Med. Chem.* **2004**, *39*, 573. (b) Condello, C.; Scharin, A.; Gruntzendler, J. *Scientific Reports* **2011**, *1*, 1.
(7) Higuchi, M.; Iwata, N.; Matsuba, Y.; Sato, K.; Sasamoto, K.; Saido, T. C. *Nat. Neurosci.* **2005**, *8*, 527.
(8) Wei, D.; Akbey, Ü.; Paaske, B.; Oschkinat, H.; Reif, B.; Bjerring, M.; Nielsen, N. C. *J. Phys. Chem. Lett.* **2011**, *2*, 1289.
(9) (a) Hermange, P.; Lindhardt, A. T.; Taaning, R. H.; Bjerglund, K.; Lupp, D.; Skrydstrup, T. *J. Am. Chem. Soc.* **2011**, *133*, 6061. (b) Hermange, P.; Gøgsig, T. M.; Lindhardt, A. T.; Taaning, R. H.; Skrydstrup, T. *Org. Lett.* **2011**, *13*, 2444. (c) Nielsen, D. U.; Taaning, R. H.; Lindhardt, A. T.; Gøgsig, T. M.; Skrydstrup, T. *Org. Lett.* **2011**, *13*, 4454. (d) Xin, Z.; Gøgsig, T. M.; Lindhardt, A. T.; Skrydstrup, T. *Org. Lett.* **2012**, *14*, 284. (e) Gøgsig, T. M.; Taaning, R. H.; Lindhardt, A. T.; Skrydstrup, T. *Angew. Chem., Int. Ed.* **2012**, *124*, 822. (f) Friis, S. D.; Taaning, R. H.; Lindhardt, A. T.; Skrydstrup, T. *J. Am. Chem. Soc.* **2011**, *133*, 18114. (g) Bjerglund, K.; Lindhardt, A. T.; Skrydstrup, T. *J. Org. Chem.* **2012**, *77*, 3793.
(10) Mantel, M. L. H.; Søbjerg, L. S.; Huynh, T. H. V.; Ebran, J.-P.; Lindhardt, A. T.; Nielsen, N. C.; Skrydstrup, T. *J. Org. Chem.* **2008**, *73*, 3570.
(11) Huynh, T. V. H.; Mantel, M. L. H.; Mikkelsen, K.; Lindhardt, A. T.; Nielsen, N. C.; Otzen, D.; Skrydstrup, T. *Org. Lett.* **2009**, *11*, 999.
(12) Shirakawa, E.; Otsuka, H.; Hayashi, T. *Chem. Commun.* **2005**, *47*, 5885.
(13) COgen, [13C]-COgen and COware are commercially available. See the Supporting Information.
(14) Park, K.; Bae, G.; Moon, J.; Choe, J.; Song, K. H.; Lee, S *J. Org. Chem.* **2010**, *75*, 6244.
(15) For some recent reviews, see: (a) Brennführer, A.; Neumann, H.; Beller, M. *Angew. Chem., Int. Ed.* **2009**, *48*, 4114. (b) Barnard, C. F. J. *Organometallics* **2008**, *27*, 5402. For some relevant examples, see: (c) Watson, D. A.; Fan, X.; Buchwald, S. L. *J. Org. Chem.* **2008**, *73*, 7096. (d) Lagerlund, O.; Larhed, M. *J. Comb. Chem.* **2006**, *8*, 4. (e) Jimenez-Rodriguez, C.; Eastham, G. R.; Cole-Hamilton, D. J. *Dalton Trans.* **2005**, 1826. (f) Mägerlein, W.; Indolese, A. F.; Beller, M. *J. Organomet. Chem.* **2002**, *641*, 30. (g) Mägerlein, W.; Indolese, A. F.; Beller, M. *Angew. Chem., Int. Ed.* **2001**, *40*, 2856. (h) Ben-David, Y.; Portnoy, M.; Milstein, D. *J. Am. Chem. Soc.* **1989**, *111*, 8742.
(16) The CO release for the optimized conditions was tested in a single chamber system at atmospheric pressure. See the Supporting Information.
(17) Perrin, D.; Armarego, W. *Purification of Laboratory Chemicals*, 3rd ed.; Pergamon Press: New York, 1988.
(18) Gottlieb, H. E.; Kotlyar, V.; Nudelman, A. *J. Org. Chem.* **1997**, *62*, 7512.
(19) Bodnar, B. S.; Vogt, P. F. *J. Am. Chem. Soc.* **2009**, *74*, 2598.