Angewandte Chemie
Eine Zeitschrift der Gesellschaft Deutscher Chemiker

Supporting Information

69451 Weinheim, Germany

The Synthesis and Biological Study of New Inducible DNA Cross-linking Agents

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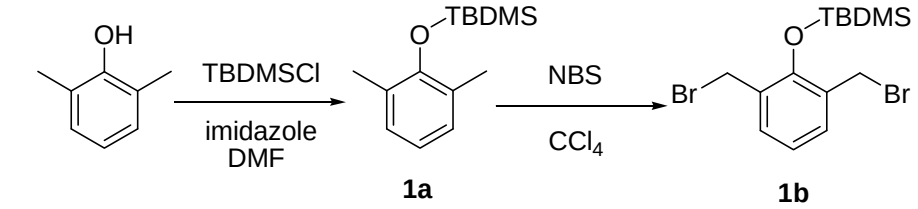
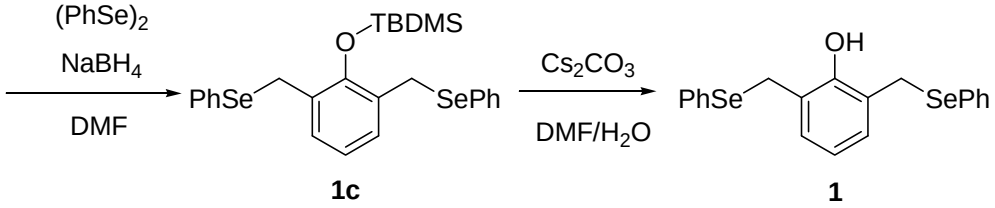
Contents:

Synthesis experiments:

General information: ^{1}H NMR (internal reference TMS) spectra were recorded on Varian Mercury 300 spectrometers. ^{13}C NMR (internal reference TMS) and ^{77}Se NMR (external reference Me_2Se) spectra were recorded on Varian Mercury 600 spectrometers. Chemical shifts were reported as δ values relative to internal standards. Mass spectra were determined using Bruker Daltonics APEXII 47e. Elemental analysis was recorded on PE-240 C.





***O*-(*t*-Butyldimethylsilyl)-2,6-dimethylphenol (1a).[1]**

2,6-Dimethylphenol (0.452 g, 3.7 mmol) was dissolved in DMF (10 mL), and imidazole (1.00 g, 14.9 mmol) and *t*-butyldimethylsilyl chloride (1.12 g, 7.4 mmol) were added The mixture was stirred at room temperature overnight. Then brine was added and the mixture was extracted with ether. The ether solution was combined, washed with brine several times, and dried with anhydrous Na_2SO_4. The solution was evaporated and the residue was subject to column chromatography on silica gel (hexane:ethyl acetate, 120:1) to give the desired product as a colorless oil (0.825 g, 94.5 % yield). ^{1}H NMR ($CDCl_3$) δ 0.22 (s, 6 H), 1.07 (s, 9 H), 2.24 (s, 6 H), 6.82 (t, J = 7.5 Hz, 1 H), 6.98 (d, J = 7.2 Hz, 2 H).

***O*-(*t*-Butyldimethylsilyl)-2, 6-bis(bromomethyl)phenol (1b).[1,2]**

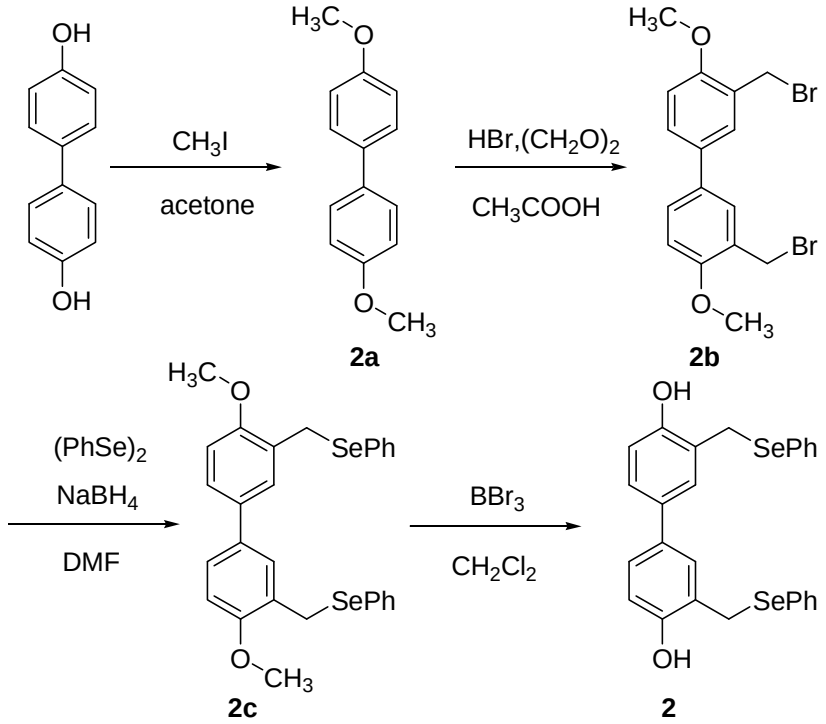
To a solution of *O*-(*t*-Butyldimethylsilyl)-2, 6-dimethylphenol (0.330 g, 1.4 mmol) in CCl_4 (20 mL) was added NBS (0.534 g, 3 mmol), and the mixture was heated to reflux. Then AIBN (13 mg) was added. The reaction was refluxed for 2 h, cooled and filtered. The CCl_4 solution was washed with water, dried with anhydrous Na_2SO_4, and concentrated under reduced pressure. The product was purified by column chromatography (hexane:ethyl acetate, 120:1) to yield a colorless oil (0.161 g, 29.2 % yield). ^{1}H NMR ($CDCl_3$) δ 0.32 (s, 6 H), 1.11 (s, 9 H), 4.52 (s, 4 H), 6.98 (t, J = 7.8 Hz, 1 H), 7.35 (d, J = 7.5 Hz, 2 H); ^{13}C NMR ($CDCl_3$) δ -3.16, 19.11, 26.30, 29.37, 122.71, 129.43, 132.51, 151.08.

***O*-(*t*-Butyldimethylsilyl)-2, 6-bis(phenylselenylmethyl)phenol (1c).[3]**

Under N_2 atmosphere, diphenyldiselenide (0.158 g, 0.508 mmol) and sodium borohydride (0.038 g, 1.016 mmol) were dissolved in dry DMF (5 mL) at room temperature for 10 min. A solution of benzyl bromide compound **1b** (0.100 g, 0.254 mmol) in DMF (10 mL) was added dropwise using syringe and stirred overnight. The mixture was diluted with ethyl acetate, washed with brine several times, dried with Na_2SO_4, and evaporated. The crude product was purified via silica gel column chromatography (hexane:ethyl acetate, 120:1) to give product as a yellow oil (0.116 g, 83.4 % yield). ^{1}H NMR ($CDCl_3$) δ 0.24 (s, 6 H), 1.04 (s, 9 H), 4.11 (s, 4 H), 6.74 (t, J = 7.5 Hz, 1H), 7.01 (d, J = 7.5 Hz, 2 H), 7.23-7.26 (m, 6 H), 7.39-7.42 (m, 4 H); ^{13}C NMR ($CDCl_3$) δ -2.98, 18.94, 26.28, 27.60, 121.85, 127.27, 129.14, 129.87, 130.07, 130.97, 133.54, 150.82; ^{77}Se NMR ($CDCl_3$) δ 363.36; Anal. Calcd for $C_{26}H_{32}OSe_2Si$: C, 57.14; H, 5.90. Found: C, 57.49; H, 5.80.

2, 6-bis(phenylselenylmethyl)phenol (1).[4]

O-(*t*-butyldimethylsilyl)-2,6-bis(phenylselenylmethyl)phenol (0.293 g, 0.535 mmol) was dissolved in DMF-H_2O (2.2 mL, V/V = 10:1). Cs_2CO_3 (0.087 g, 0.267 mmol) was added and the mixture was stirred at room temperature overnight. The solution was diluted with ethyl acetate, and washed with brine several times. Then the organic solution was dried with anhydrous Na_2SO_4, and concentrated under reduced pressure. The residue was purified by column chromatography over silica gel (hexane:ethyl acetate, 60: 1) to yield the desired product as a yellow oil (0.160 g, 69.3 % yield). ^{1}H NMR ($CDCl_3$) δ 4.15 (s, 4 H), 6.22 (s, 2 H), 6.61 (t, *J* = 7.2 Hz, 1 H), 6.83 (d, *J* = 7.2 Hz, 2 H), 7.20-7.26 (m, 6 H), 7.44-7.47 (m, 4 H); ^{13}C NMR ($CDCl_3$) δ 28.13, 120.55, 125.40, 127.89, 129.25, 129.60, 129.92, 134.31, 152.71; ^{77}Se NMR ($CDCl_3$) δ 327.44; HRMS (M^+) calc.433.9688, Found 433.9687.



4, 4'-Dimethoxy-1,1'-biphenyl (2a).

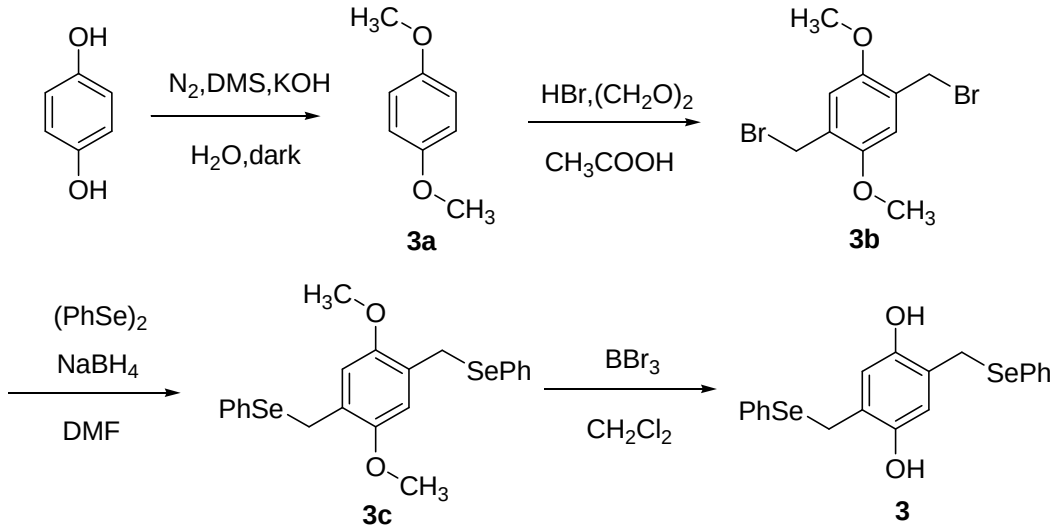
Commercially available 4,4'-dihydroxy-1,1'-biphenyl (1.86 g, 10 mmol) was dissolved in 100 mL acetone and 10 mL N,N-dimethylformamide. Then solid carbonate potassium (6.90 g, 50 mmol) and excessive iodomethane (5 mL, 49 mmol) liquid were added and the mixture was stirred at room temperature until the reaction was finished as being indicated by thin-layer chromatography (TLC). The mixture was then concentrated under reduced pressure to remove the excessive iodomethane. The residue was separated by chloroform and water. The organic phase was collected, dried with anhydrous Na_2SO_4, and evaporated to give the product as white solid (1.90 g, 88.8 % yield). ^{1}H NMR ($CDCl_3$) δ 3.85 (s, 6 H), 6.95 (d, *J* = 8.7 Hz, 4 H), 7.46 (d, *J* = 9.0 Hz, 4 H).

3,3'-Di(bromomethyl)-4,4'-dimethoxy-1,1'-biphenyl (2b). [5]

Paraformaldehyde (1.332 g, 44.4 mmol) and **2a** (1.90 g, 8.88 mmol) were suspended in 50 mL acetic acid and 5 mL 40 % HBr in 30 mL acetic acid was added to the mixture in one portion at 80 ^{o}C. The suspensive solution became clear immediately. The reaction was finished after 3 h at this temperature, cooled and crystallized for 24 h at 4 ^{o}C. The crystal was filtered, washed with water, and dissolved in dichloromethane. The organic phase was washed by saturated sodium bicarbonate aqueous solution several times, dried with anhydrous Na_2SO_4 and evaporated. The residue was purified by column chromatography (hexane:ethyl acetate, 10:1) to yield the desired product as white solid (0.92 g, 30 % yield). ^{1}H NMR ($CDCl_3$) δ 3.94 (s, 6 H), 4.62 (s, 4 H), 6.93 (d, *J* = 8.4 Hz, 2 H), 7.45 (dd, *J* = 2.4 Hz, *J* = 8.4 Hz, 2 H), 7.51 (d, *J* = 2.40 Hz, 2 H); ^{13}C NMR ($CDCl_3$) δ 29.19, 56.02, 111.56, 126.65, 128.52, 129.44, 133.17, 156.92. FAB-MS (M^+) 400.

3,3'-bis(phenylselenylmethyl)-4,4'-dimethoxy-1,1'-biphenyl (2c).
This reaction was carried out following the same procedure as the synthesis of **1c** .The product was purified via silica gel column chromatography (chloroform:hexane, 2:3) to give product as a white solid.(0.343 g, 62 % yield). ^{1}H NMR ($CDCl_3$) δ 3.84 (s, 6 H), 4.12 (s, 4 H), 6.83 (d, *J* = 8.4 Hz, 2 H), 6.92 (d, *J* = 2.1 Hz, 2 H), 7.21-7.25 (m, 8 H), 7.48-7.52 (m, 4 H); ^{13}C NMR ($CDCl_3$) δ 27.31, 55.80, 110.98, 126.42, 127.51, 127.72, 128.74, 129.07, 130.93, 132.90, 134.52, 156.33; ^{77}Se NMR ($CDCl_3$) δ 374.44; Anal. Calcd for $C_{28}H_{26}O_2Se_2$: C, 60.88; H, 4.74. Found: C, 60.50; H, 4.60.

3, 3'-bis(phenylselenylmethyl)-4,4'-dihydroxy-1,1'-biphenyl (2). [6]
Under N_2 atmosphere, at 0 ^{o}C, BBr_3 (neat, 0.26 mL, 2.88 mmol) was added gradually to a solution of **2c** (0.2 g, 0.36 mmol) in fresh distilled CH_2Cl_2 (20 mL). After the mixture was stirred for 2 h at room temperature, the solution was poured into a large amount of ice-water and extracted with $CHCl_3$. The organic phase was washed with brine, dried over anhydrous Na_2SO_4, and evaporated. The residue was purified by column chromatography (hexane:ethyl acetate, 3:1) to give the desired product as a white solid (0.089 g, 46.7 % yield). ^{1}H NMR ($CDCl_3$) δ 4.13 (s, 4 H), 5.53 (s, 2 H), 6.81-6.85 (m, 4 H), 7.12 (dd, *J* = 2.4 Hz, *J* = 8.4 Hz, 2 H), 7.23-7.27 (m, 6 H), 7.46-7.49 (m, 4 H). ^{13}C NMR ($CDCl_3$) δ 28.08, 116.96, 124.67, 127.13, 128.04, 129.08, 129.30, 129.50, 133.54, 134.60, 153.25; ^{77}Se NMR ($CDCl_3$) δ 333.83; Anal. Calcd for $C_{26}H_{22}O_2Se_2$: C, 59.55; H, 4.23. Found: C, 59.68; H, 4.50.



1, 4-dimethoxybenzene (3a).[7]
Dimethyl sulfate (2.84 mL, 30 mmol) and potassium hydroxide (1.12 g, 20 mmol) was added to the solution of hydroquinone (1.1 g, 10 mmol) in 100 mL water under N_2 atmosphere and dark condition. Then the mixture was stirred at room temperature for 8 h. The precipitation in the solution was filtered, washed with water several times, and dried in vacuum to give the bis-methyl ether as white solid (1.045 g, 75.7 % yield). ^{1}H NMR ($CDCl_3$) δ 3.78 (s, 6 H), 6.85 (s, 4 H)

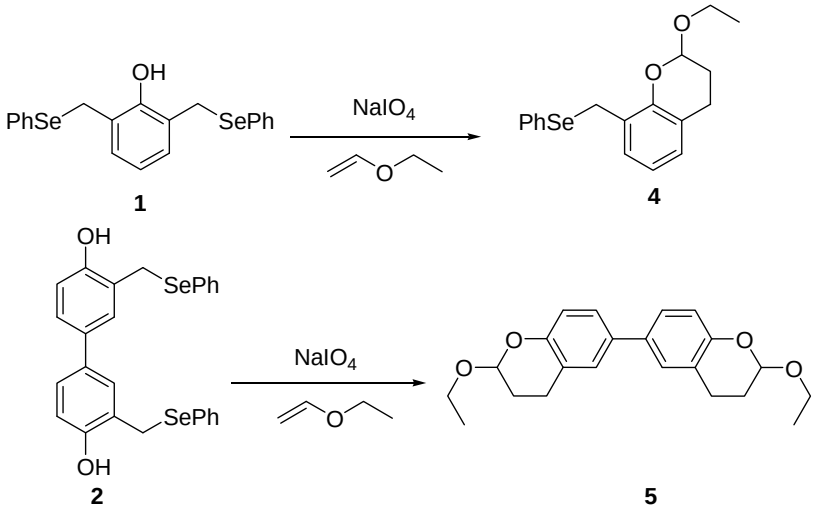
1, 4-bis(bromomethyl)-2, 5-dimethoxybenzene (3b).
3b was synthesized following the same procedure as the synthesis of **2b**. Paraformaldehyde (1.087 g, 36.25 mmol) and **3a** (1 g, 7.25 mmol) were suspended in 50 mL acetic acid and 5 mL 40 % HBr in 30 mL acetic acid was added to the mixture in one portion at 80 ^{o}C. The suspensive solution became clear immediately. The reaction was finished after 3 h at this temperature, cooled and crystallized for 24 h at 4 ^{o}C. The crystal was filtered, washed with water, and dried in vacuum to give the dibromo product as white solid (1.630 g, 69.4 % yield). ^{1}H NMR ($CDCl_3$) δ 3.87 (s, 6 H), 4.53 (s, 4 H), 6.87 (s, 2 H); ^{13}C NMR ($CDCl_3$) δ 28.79, 56.48, 114.08, 127.66, 151.51; FAB-MS (M^+) 322.

1, 4-bis(phenylselenylmethyl)-2,5-dimethoxybenzene (3c).

3c was synthesized following the same procedure as the synthesis of **1c.** The crude product was purified via silica gel column chromatography (chloroform:hexane, 1:1) to give product (0.290 g, 82.6 % yield). ^{1}H NMR ($CDCl_3$) δ 3.58 (s, 6 H), 4.05 (s, 4 H), 6.41 (s, 2 H), 7.22-7.25 (m, 6 H), 7.45-7.47 (m, 4 H); ^{13}C NMR ($CDCl_3$) δ 27.13, 56.16, 113.36, 126.88, 127.44, 129.02, 130.86, 134.41, 150.70; ^{77}Se NMR ($CDCl_3$) δ 375.93; Anal. Calcd for $C_{22}H_{22}O_2Se_2$: C, 55.47; H, 4.66. Found: C, 55.41; H, 4.75.

2, 5-bis(phenylselenylmethyl)benzene-1,4-diol (3).

Compound **3** was synthesized following the same procedure as the synthesis of **2** using neat BBr_3. The crude product was purified by column chromatography (hexane:ethyl acetate, 4: 1) to give the desired product. (0.136 g, 50 % yield).^{1}H NMR ($CDCl_3$) δ 4.01 (s, 4 H), 5.03 (s, 2 H), 6.45 (s, 2 H), 7.22-7.27 (m, 6 H), 7.43-7.46 (m, 4 H); ^{13}C NMR ($CDCl_3$) δ 27.82, 118.67, 124.95, 128.03, 129.30, 134.23, 147.92; ^{77}Se NMR ($CDCl_3$) δ 329.94; Anal. Calcd for $C_{20}H_{18}O_2Se_2$: C, 53.59; H, 4.05. Found: C, 55.27; H, 4.28.



The trap reactions:

A solution of substrate (0.05 g) in 5 mL acetronitrile-H_2O (V/V = 3:2) in presence of excess ethyl vinyl ether was incubated at 37 ^{o}C for 0.5 h. Then $NaIO_4$ was added to start the reaction for 3 h at the same temperature. The residue was separated by ethyl acetate and water. The organic phase was collected, dried with anhydrous Na_2SO_4, and evaporated. The residue was purified by column chromatography (hexane:ethyl acetate, 120:1) to give the desired product.

Compound **4**: brown oil, 0.018 g, 44.7 % yield. ^{1}H NMR ($CDCl_3$) δ 1.15 (dd, *J* = 7.5 Hz, J = 6.6 Hz, 3 H), 1.87-2.01 (m, 2 H), 2.53-2.61 (m, 1 H), 2.85-2.96 (m, 1 H), 3.57-3.63 (m, 1 H), 3.90-3.96 (m, 1 H), 4.07 (s, 2 H), 5.22 (s, 1 H), 6.67 (dd, *J* = 7.5 Hz, *J* = 7.2 Hz, 1 H), 6.85 (dd, *J* = 8.7 Hz, *J* = 8.1 Hz, 2 H), 7.17-7.19 (m, 3 H), 7.43-7.44 (m, 2 H); ^{13}C NMR ($CDCl_3$) δ 15.32, 20.97, 26.77, 27.10, 64.08, 97.27, 120.16, 122.79, 126.74, 127.20, 128.24, 128.46, 129.03, 131.60, 133.74, 150.06; ^{77}Se NMR ($CDCl_3$) δ 363.36; Anal. Calcd for $C_{18}H_{20}O_2Se$: C, 62.25; H, 5.80. Found: C, 62.30; H, 5.98.

Compound **5**: brown oil, 0.012 g, 34.3 % yield. ^{1}H NMR ($CDCl_3$) δ 1.14 (t, *J* = 7.5 Hz, *J* = 6.6 Hz, 6 H), 1.87-2.03 (m, 4 H), 2.57-2.65 (m, 2 H), 2.89-3.01 (m, 2 H), 3.53-3.60 (m, 2 H), 3.79-3.87 (m, 2 H), 5.20 (s, 4 H), 6.77-6.80 (d, *J* = 8.1 Hz, 2 H), 7.15-7.22 (m, 4 H), ^{13}C NMR ($CDCl_3$) δ 15.35, 20.95, 26.87, 63.93, 97.26, 117.35, 122.89, 125.91, 127.74, 133.87, 151.57; HRMS (M^+) calc. 354.1826. Found 354.1828.

DNA experiments:

General Information: Plasmid DNA (pBR322) was purchased from TOYOBO Co., Ltd.. The DNA oligonucleotide was purchased from Invitrogen Biotechnology Co.Ltd., with the 5′-terminus fluoro-labeled with TAMRA. The 39-mer oligonucleotide used in Figure 3 and S3 with the following sequence

5'-CATGG TGGTT TGGGT TAGGG TTAGG GTTAG GGTTA CCAC-3'. The molecular weight of this oligo is 12196.8.

General procedure for linearization of plasmid pBR322 by EcoR I:
Supercoiled pBR322 (10 μL,10 μg) was incubated with EcoR I (10 μL, 40-80 u), EcoR I buffer (20 μL), acetylated BSA (20 μL, 1 mg/mL) and 140 μL H_2O (sterile) for 3 h at 37 ^oC. NaOAc (20 μL, 3 M) and ethanol (750 μL) were added, and the solution was cooled at -20 ^oC overnight. The mixture was centrifuged for 10 min at 16000 rpm, and the ethanol was decanted off. The remaindered ethanol was evaporated at -20 ^oC. The remaining linearized DNA was solved in 50 μL sterile H_2O.

General protocol for alkaline agarose gel electrophoresis:
See reference 8.

Cross-linking of DNA oligonucleotide by compounds:
Cross-linking reaction was carried out in a volume of 10 μL containing 8 μM selective 5′-terminus fluoro-labeled oligonucleotide, 10 mM phosphate buffer (pH = 7.7) and 10 mM compound. The mixture was incubated at 37 ^oC for 3 h. Then $NaIO_4$ aqueous solution was added at the final concentration of 50 mM. After 1 h, the solution was mixed with 10 μL formamide deionized to the final volume of 20 μL. Half of the residue was took out and then analyzed by a 20 % denaturing polyacrylamide electrophoresis. The cross-linking products of compound **1** and **2** with oligonucleotide were produced follow the same protocol mentioned as above.

Piperidine treatment and Fe(Ⅱ)•EDTA cleavage:
The cross-linked products were isolated by electrophoresis and determined the site of alkylation by various treatments to induce strand scission. The cross-linked product (10 pmol) was treated with 100 μL 1 M piperidine for 30 min at 90 ^oC. After precipitated by ethanol at –20 ^oC for one night. The DNA product fragments were then dissolved in formamide deionized, analyzed by polyacrylamide gel electrophoresis under denaturing conditions (7 M urea). The Fe(Ⅱ)•EDTA cleavage reactions were conducted on gel-purified cross-linked DNA (10 pmol) as previously described by Hopkins.[9] DNA was treated with a solution of 500 μM $(NH_4)_2Fe(SO_4)_2$, 1 mM EDTA, 10 mM sodium ascorbate, 0.1 M H_2O_2, 10 mM phosphate buffer (pH = 7.7) in total volume of 10 μL for 1 min at room temperature, and quenched by the addition of 1uL of 0.1 M thiourea. Samples were lyophilized and analyzed by DPAGE. The polyacrylamide gel was scanned by typhoon 9200 with Fluorescence mode. The preparation of DNA Maxam-Gilbert G and G + A ladder was followed with the standard methods.[10]

Reference:

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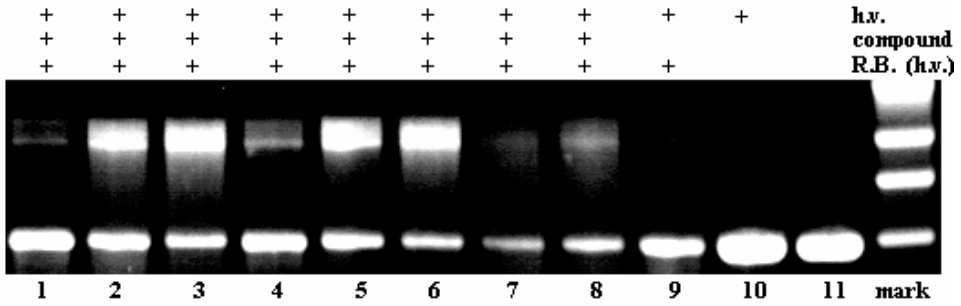
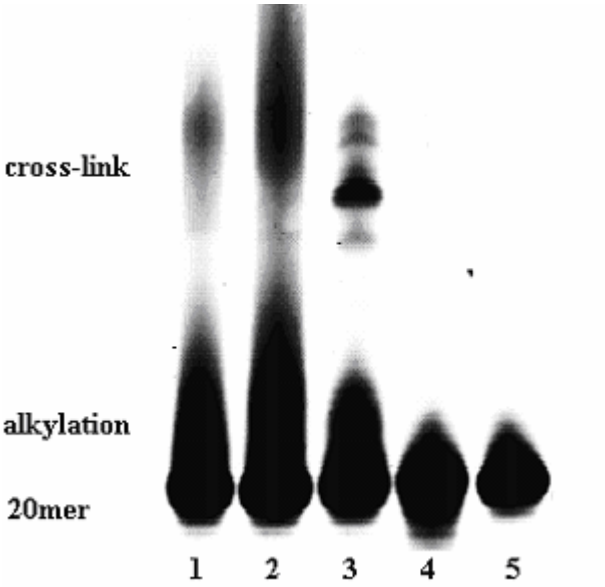


Supporting Information Figure S1: Concentration dependence of compounds **1-3** for DNA cross-linking (rose Bengal + photolysis).
Lane 1-9. with rose bengal (10 μM) + hv (1 h).
Lane 1, 0.7 μg pBR322 + 10 μM **1** (cross-linking yield 8%); lane 2, 0.7 μg pBR322 + 100 μM **1** (60%); lane 3, 0.7 μg pBR322 + 1000 μM **1** (72%); lane 4, 0.7 μg pBR322 + 10 μM **2** (32%); lane 5, 0.7 μg pBR322 + 100μM **2** (66%); lane 6, 0.7 μg pBR322 + 200 μM **2** (77%); lane 7, 0.7 μg pBR322 + 100 μM **3** (27%); lane 8, 0.7 μg pBR322 + 1000 μM **3** (54%). lane 9, 0.7 μg pBR322 + rose bengal; lane 10, 0.7 μg pBR322 + hv (hv, control); lane 11, 0.7 μg pBR322 (DNA control); Mark lane, 1.5 μg lambda DNA/HindIII (molecular weight standard)

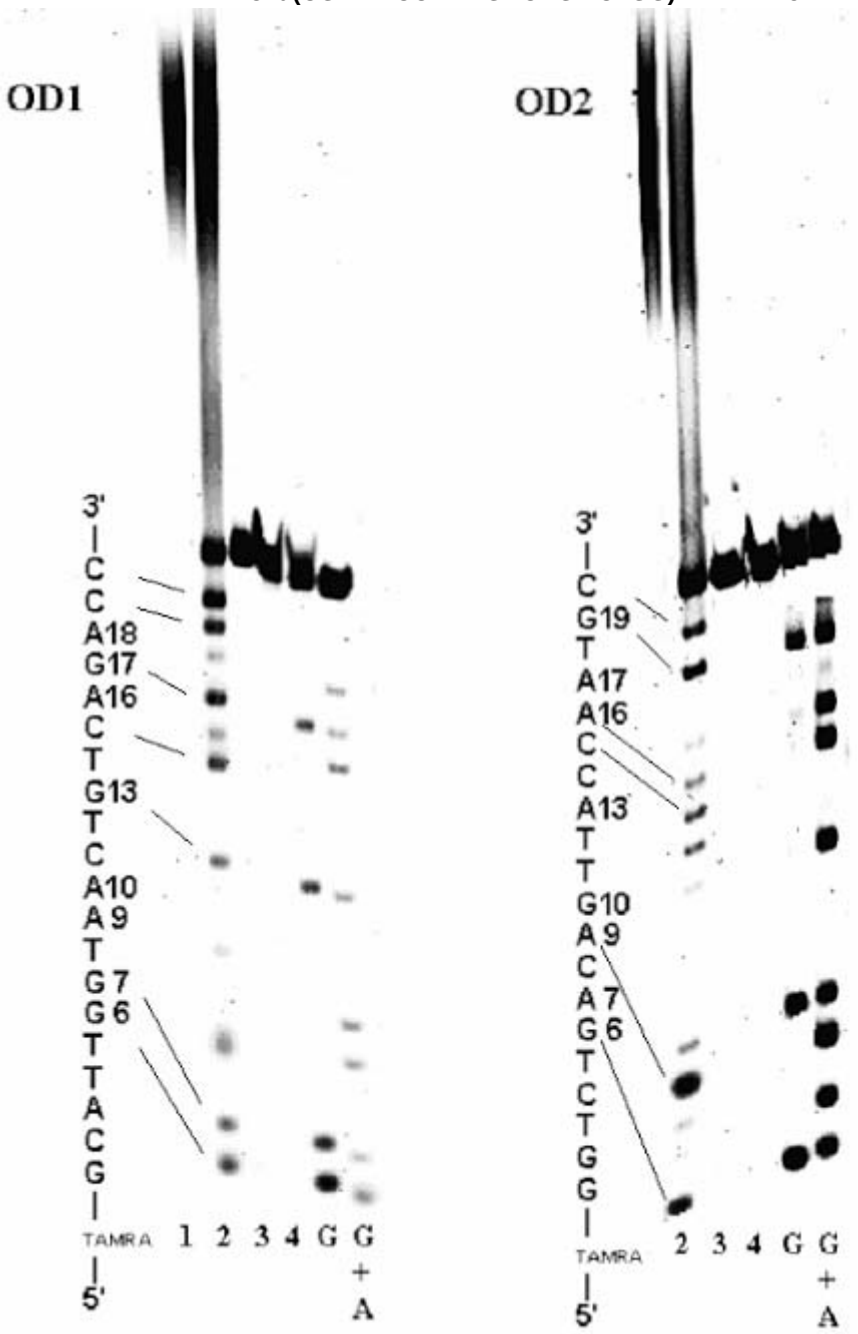


Supporting Information Figure S2: The cross-linking of oligonucleotide with compound **1**, **2** with Nitrogen mustard ISC marker.
Lane 1: compound 1 (10 mM) + oligo (4 μM) + $NaIO_4$ (50 mM) (cross-linking yield 9 %)
Lane 2: compound 2 (10 mM) + oligo (4 μM) + $NaIO_4$ (50 mM) (cross-linking yield 13 %)
Lane 3: Nitrogen mustard (10 mM) + oligo (4 μM) (cross-linking yield 12 %)
Lane 4: oligo (4 μM) + $NaIO_4$ (50 mM)
Lane 5: 20-mer oligo (4 μM)

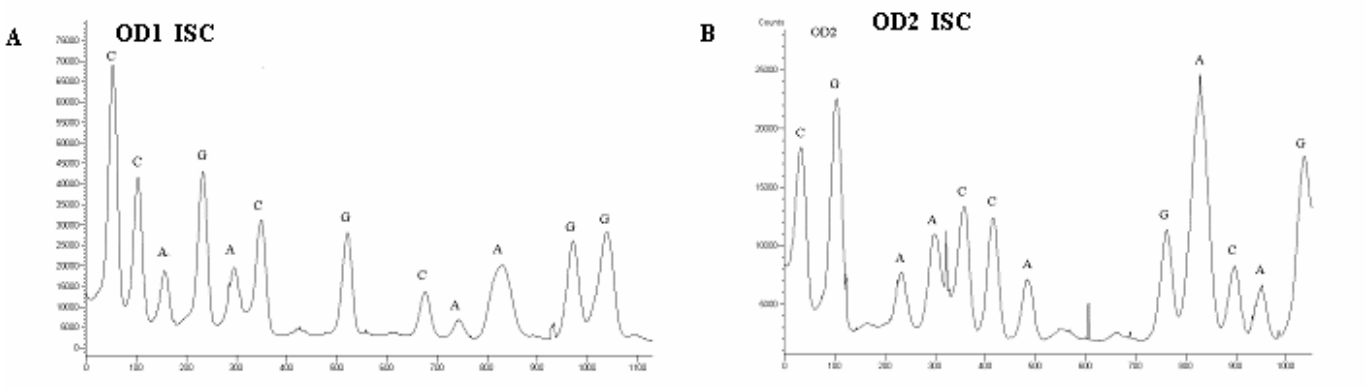
Supporting Information Figure S3: The cross-linking of compound **1**, **2** with 39-mer oligo marker.
Lane 1: 39-mer oligo
Lane 2: compound 1 (10 mM) + oligo (4 μM) + $NaIO_4$ (50 mM) (cross-linking yield 6 %)
Lane 3: compound 2 (10 mM) + oligo (4 μM) + $NaIO_4$ (50 mM) (cross-linking yield 10 %)
Lane 4: 20-mer oligo (4 μM)

OD1 5'-TAMRA-d(GCATT GGTAA CTGTC AGACC)-3'
3' -d(CGTAA CCATT GACAG TCTGG)-5'

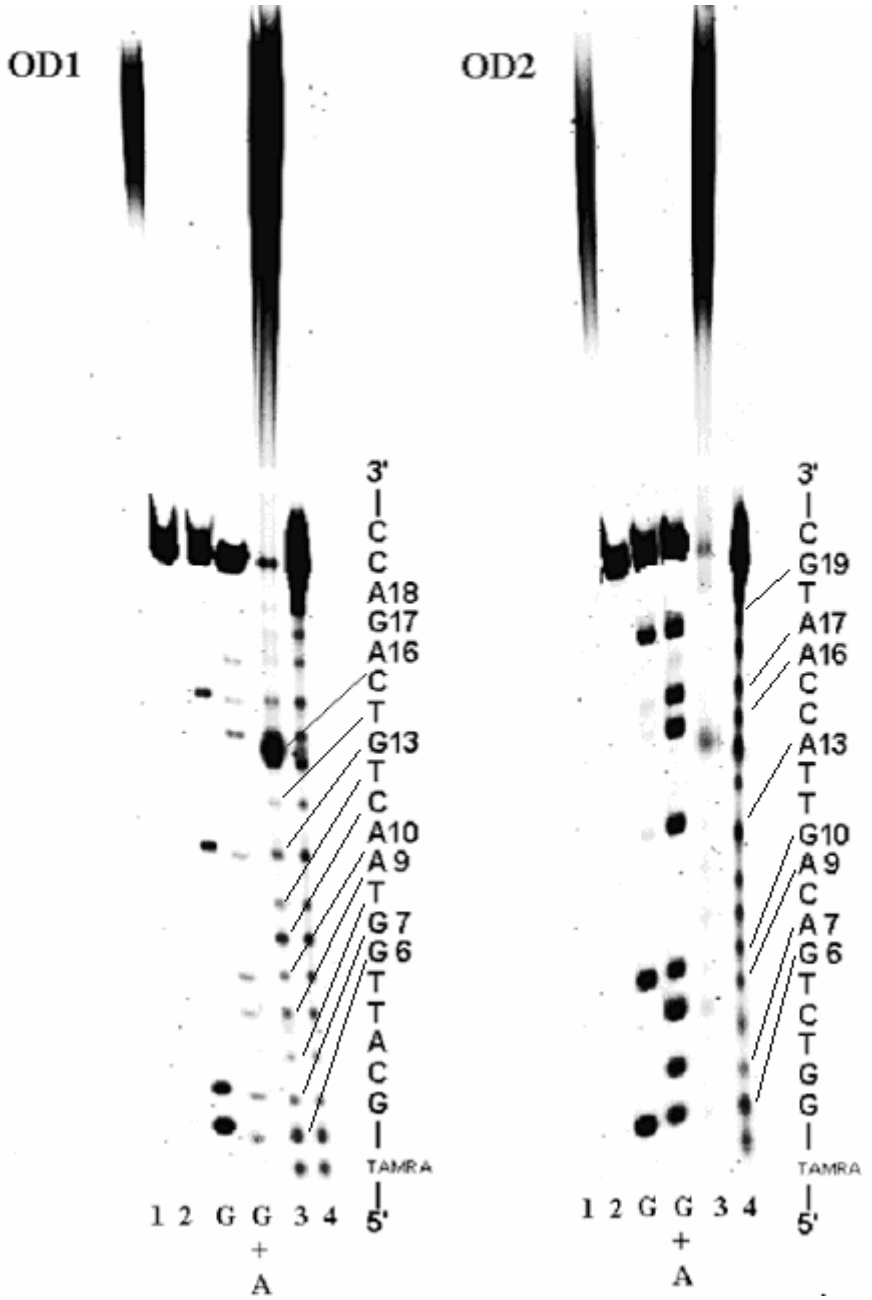
OD2 5'-d(GCATT GGTAA CTGTC AGACC)- 3'
3'-d(CGTAA CCATT GACAG TCTGG)-TAMRA- 5'



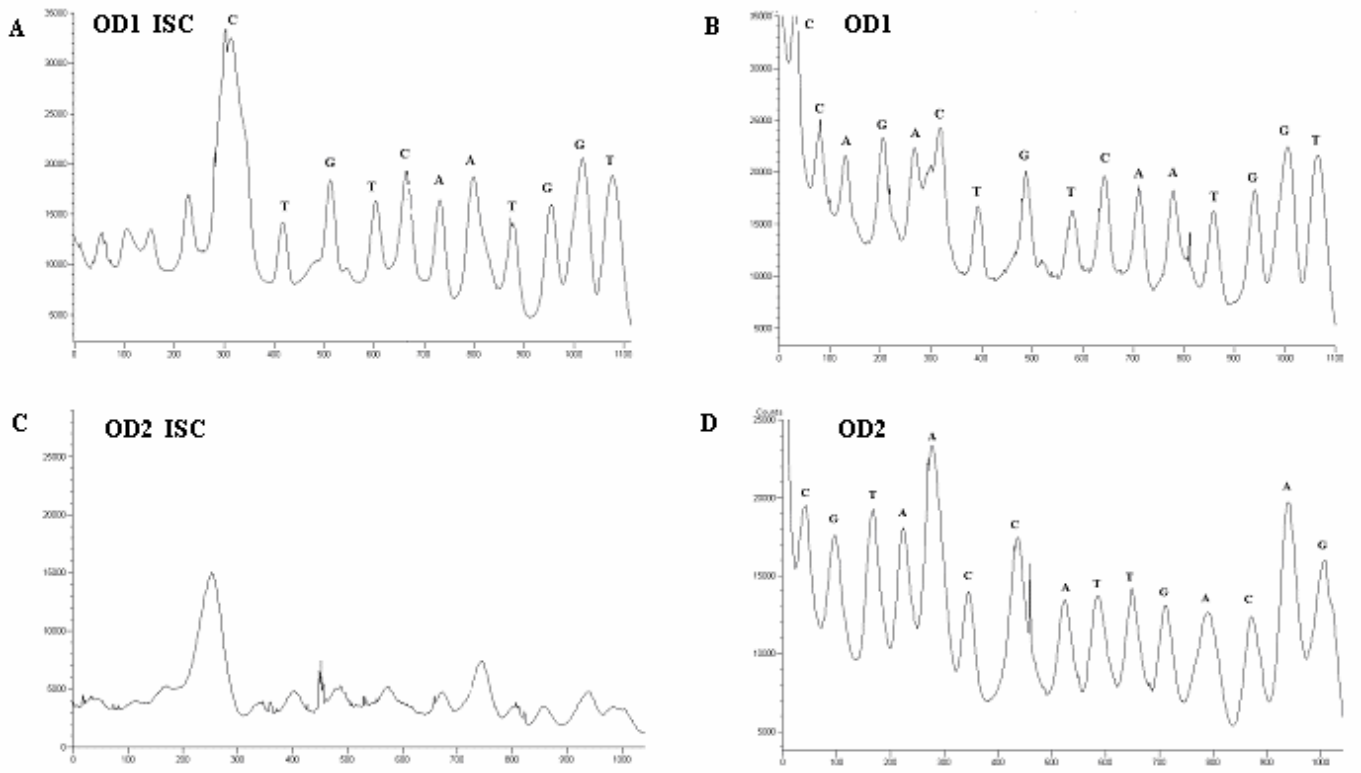
Supporting Information Figure S4: The cross-linked products of compound **2** and the piperidine treatment.
Lane1: The cross-linked products isolated from gel after gel electrophoresis.
Lane2: The isolated cross-linked product incubated with piperidine. (1 M, 90 °C, 30 min).
Lane3: uncross-linked OD1 or OD2 with piperidine.
Lane4: OD1 or OD2 control
Maxam-Gilbert G ladder
Maxam-Gilbert G + A ladder



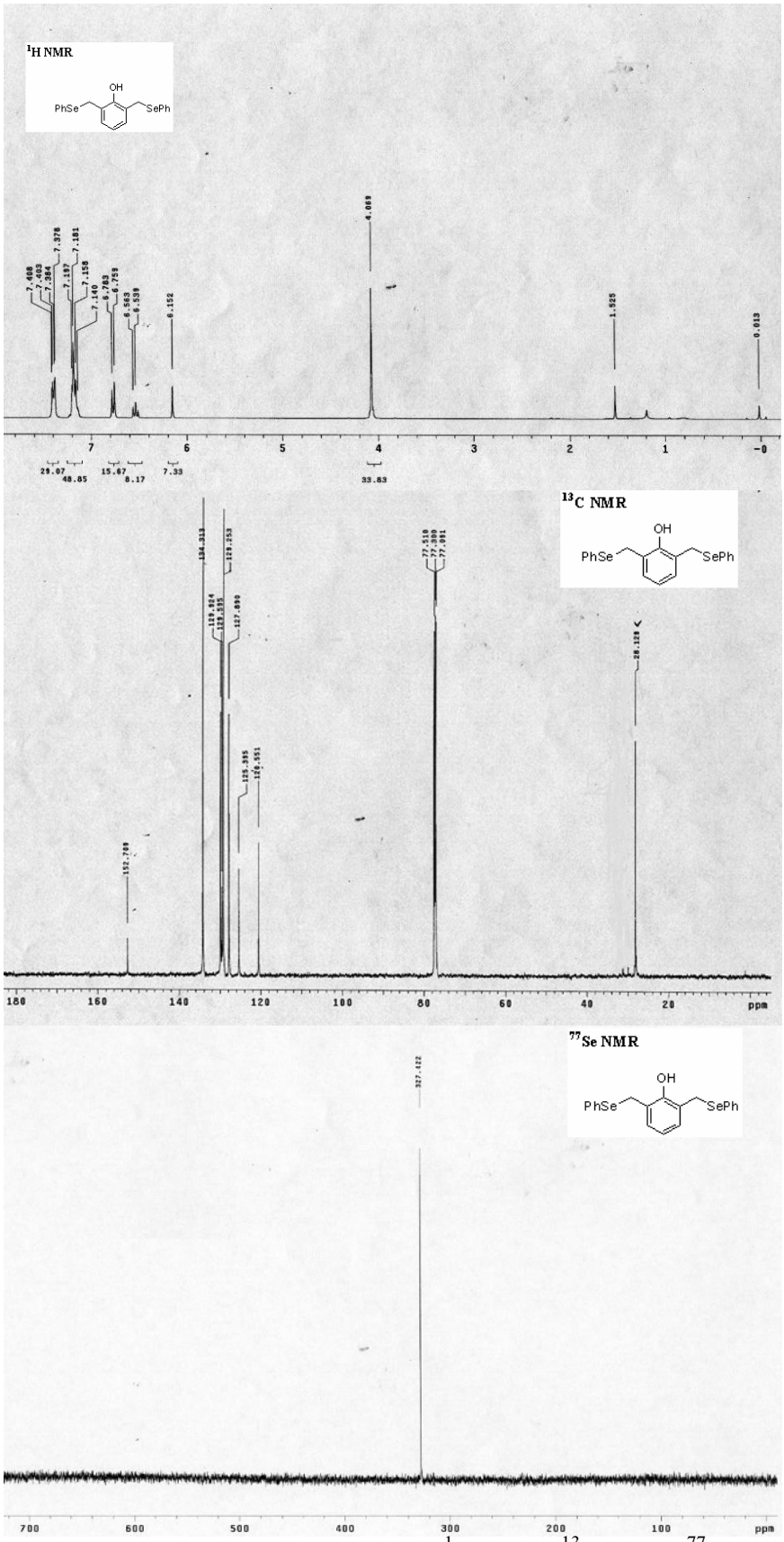
Supporting Information Figure S5: Densitometer scans of lane 2 of the DPAGE shown in Supporting Information Figure S3
A) Fragments generated by piperidine of ISC product of OD1 and compound **2**
A) Fragments generated by piperidine of ISC product of OD2 and compound **2**



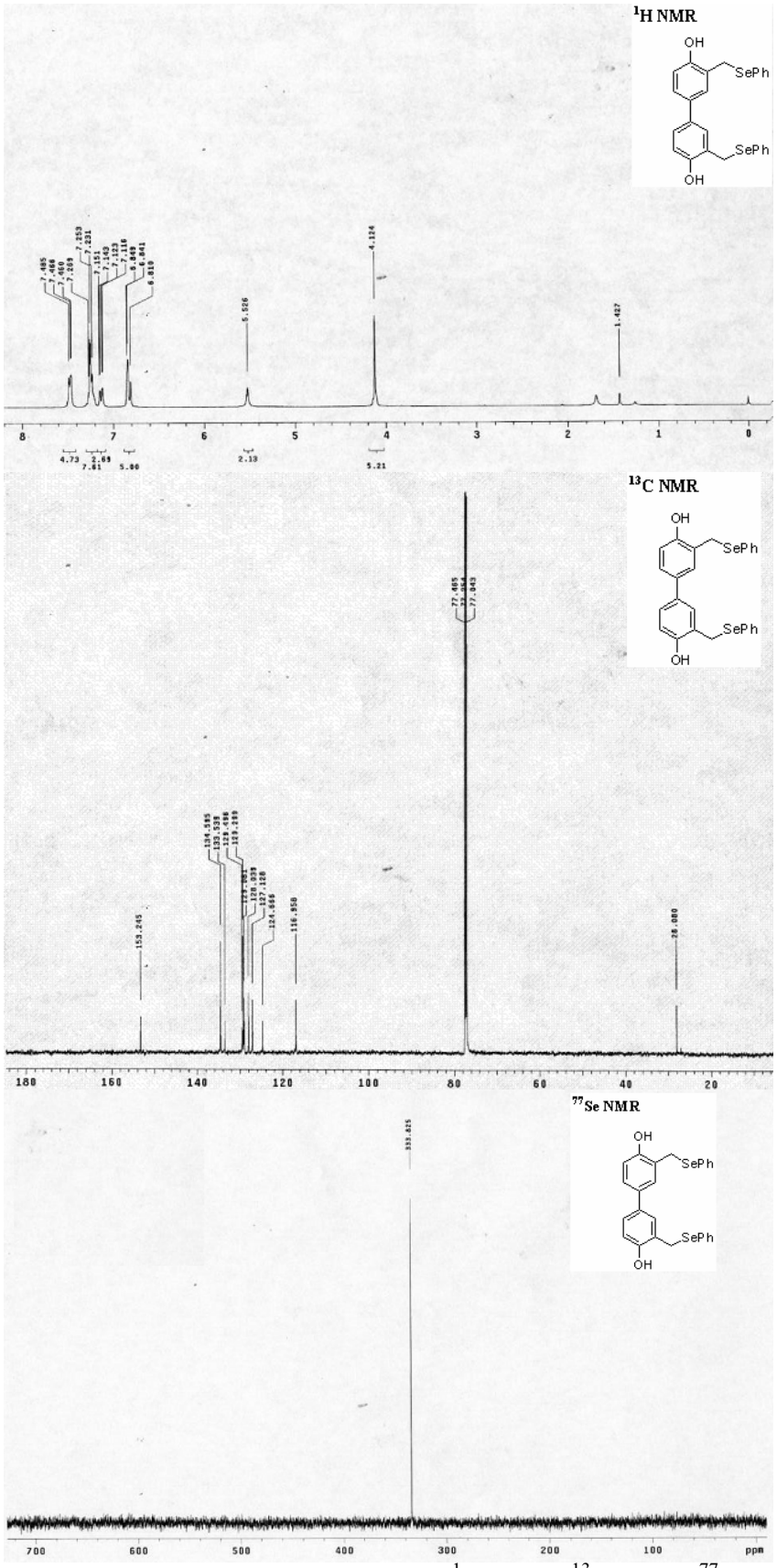
Supporting Information Figure S6: Characterization of Fe•EDTA cleavage of cross-linked product from compound **2.**
Lane1: The cross-linked products isolated from gel after gel electrophoresis.
Lane2: OD1 or OD2 control
Maxam-Gilbert G ladder
Maxam-Gilbert G + A ladder
Lane3: The Fe•EDTA treatment of isolated cross-linked products
Lane4: The Fe•EDTA treatment of uncross-linked OD1 and OD2



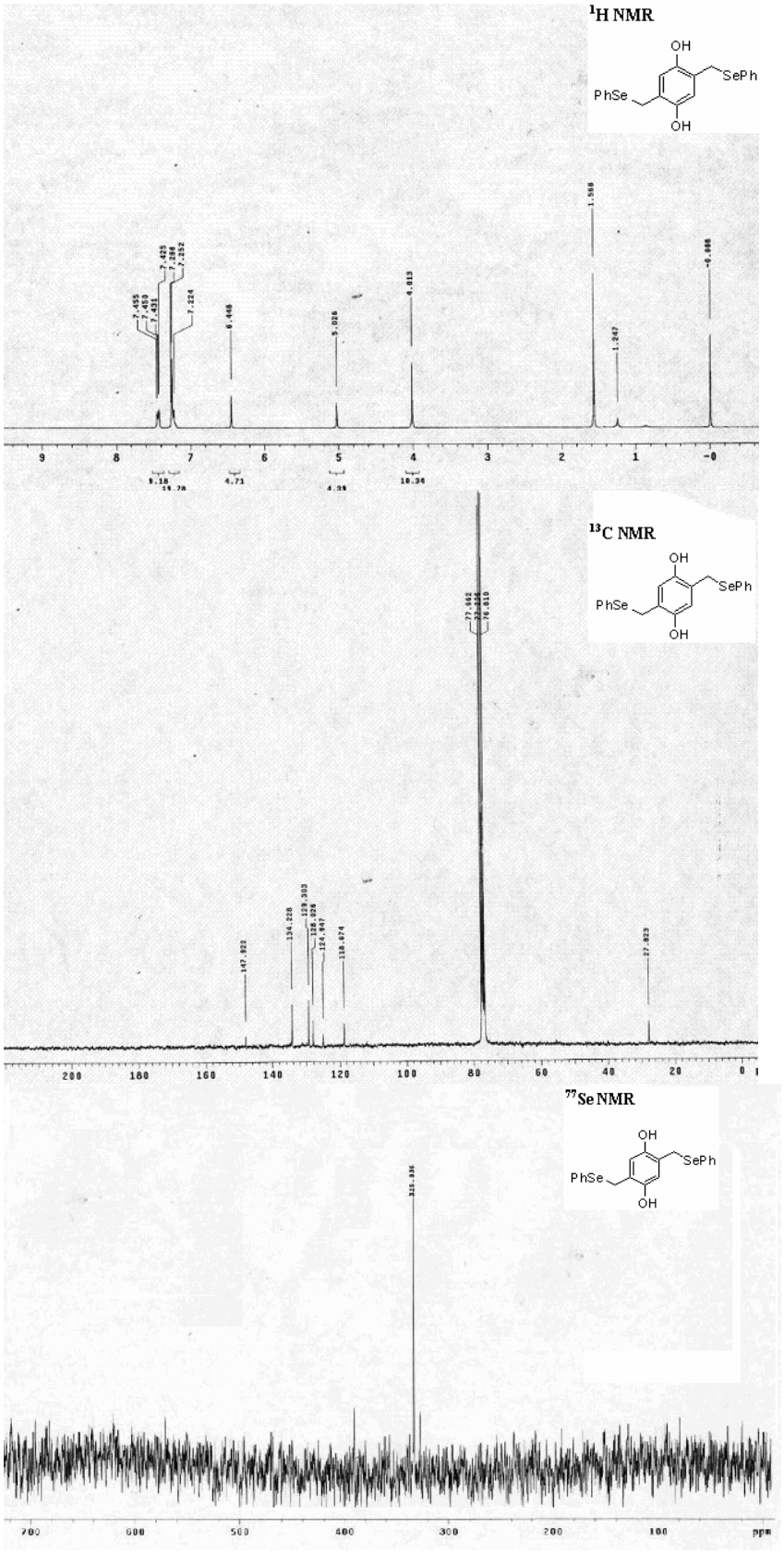
Supporting Information Figure S7: Densitometric scan of lane 3 and lane 4 of the DPAGE shown in Supporting Information Figure S4.
A) The Fe•EDTA treatment of cross-linked products of OD1 and compound **2**.
B) The Fe•EDTA treatment of OD1
C) The Fe•EDTA treatment of cross-linked products of OD2 and compound **2**
D) The Fe•EDTA treatment of OD2



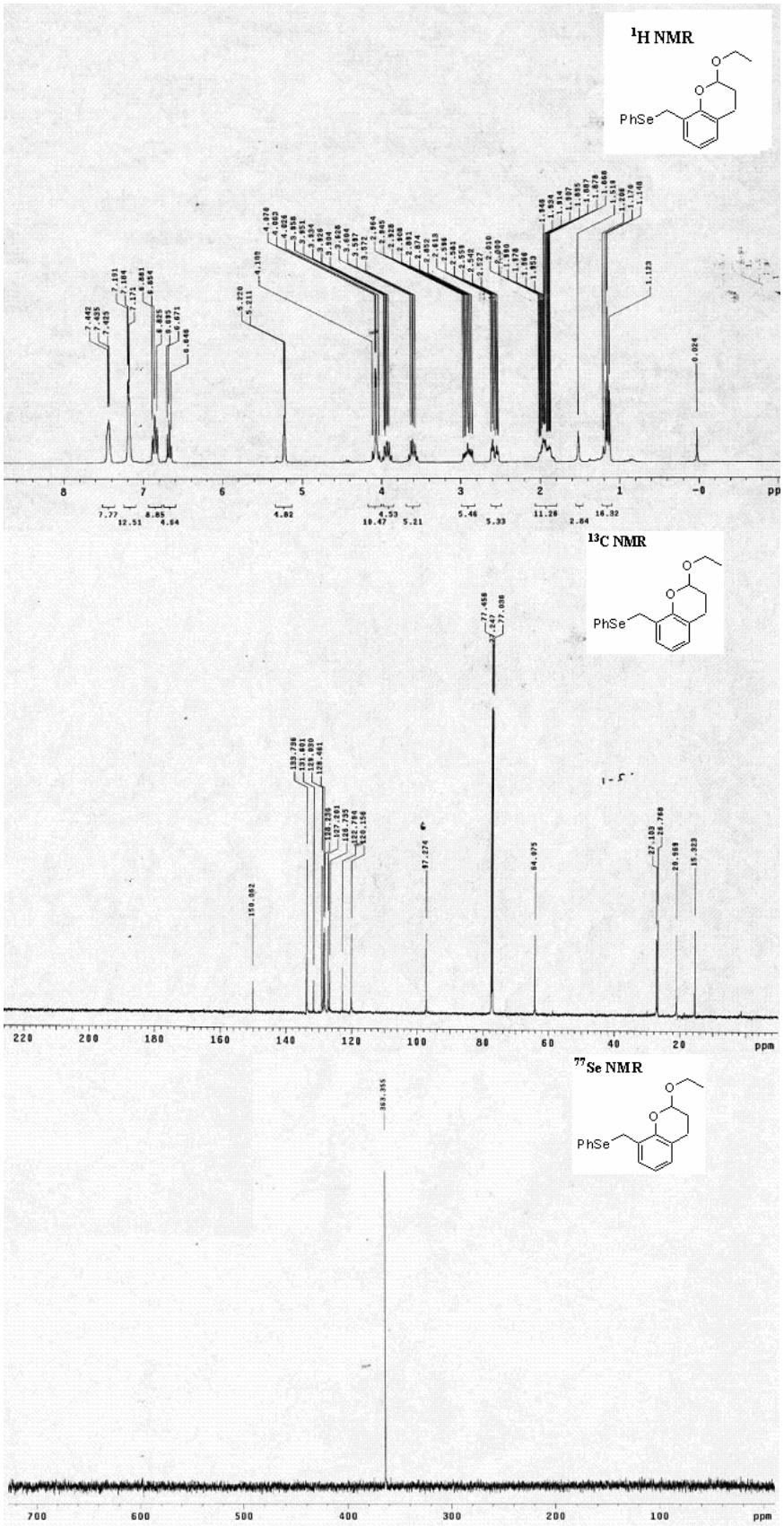
Supporting Information Figure S8: ^{1}H NMR, ^{13}C NMR, ^{77}Se NMR spectra of **1**



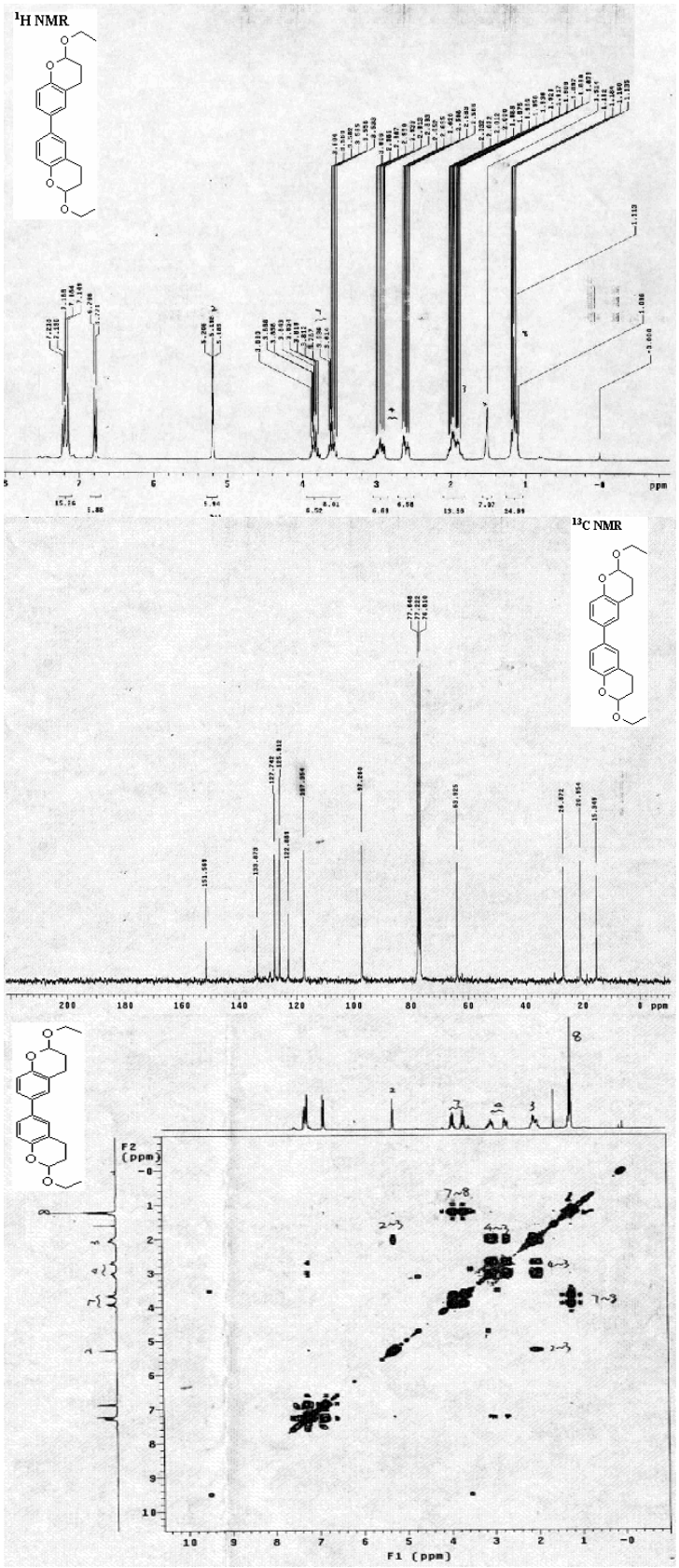
Supporting Information Figure S9: ^{1}H NMR, ^{13}C NMR, ^{77}Se NMR spectra of **2**



Supporting Information Figure S10: ^{1}H NMR, ^{13}C NMR, ^{77}Se NMR spectra of **3**

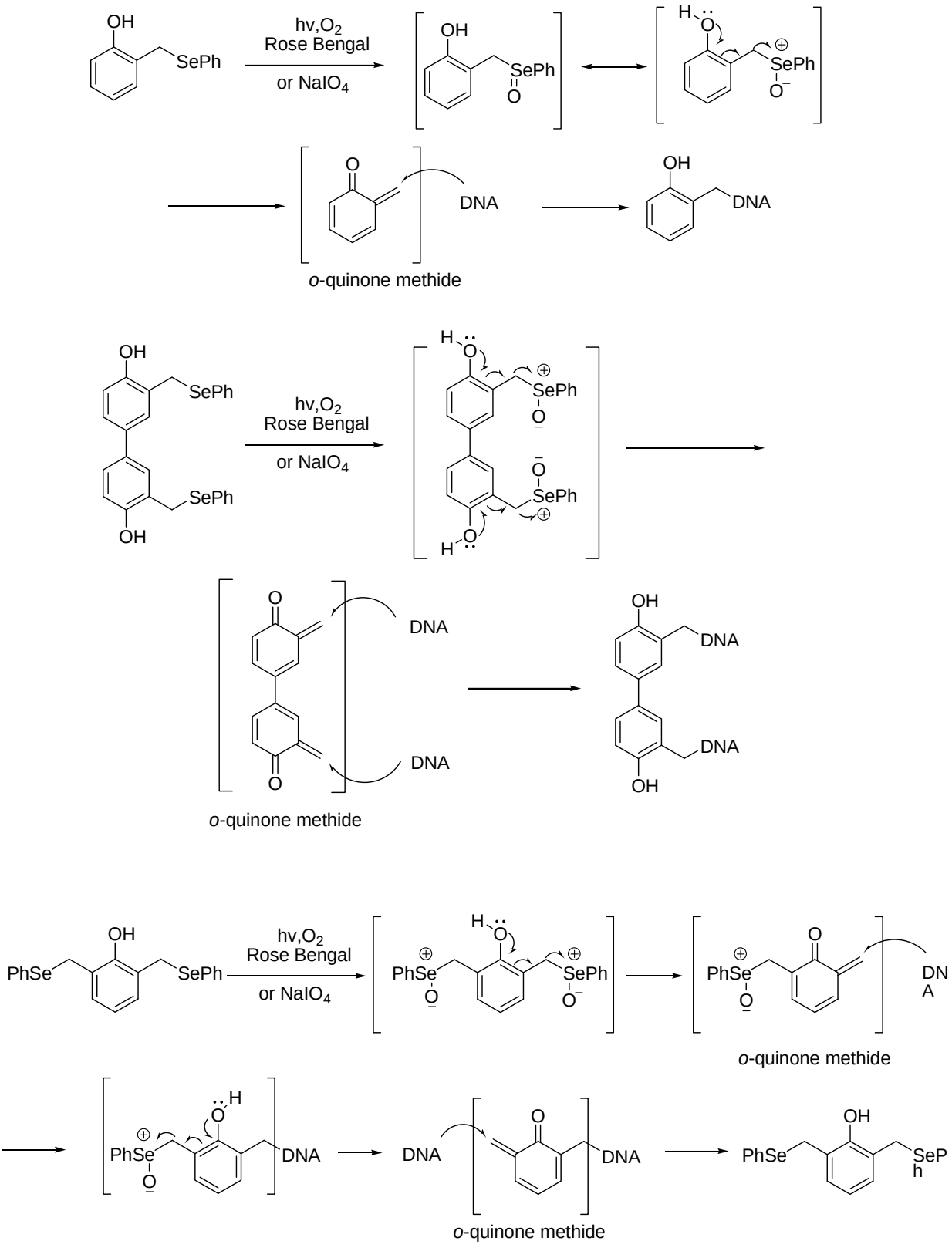


Supporting Information Figure S11: ^{1}H NMR, ^{13}C NMR, ^{77}Se NMR spectra of **4**

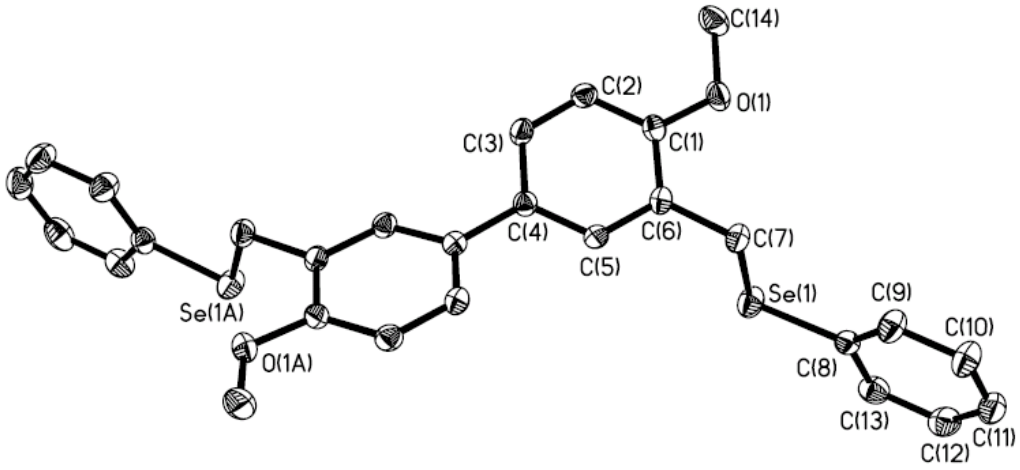


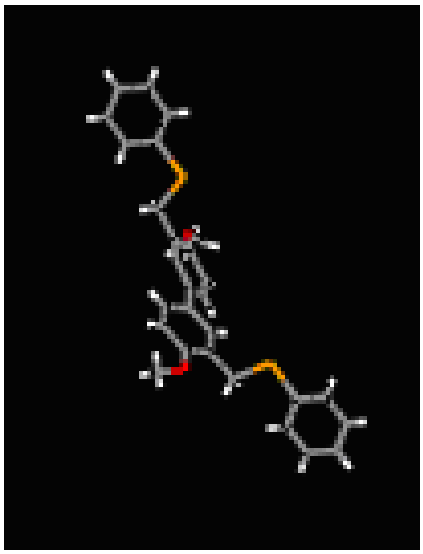
Supporting Information Figure S12: ^{1}H NMR, ^{13}C NMR, H, H-gCOSY spectra of **5**

Supporting Information Scheme S1: The formation of *o*-quinone methide intermediate.



X-Ray Crystal Structure Information for 2C.





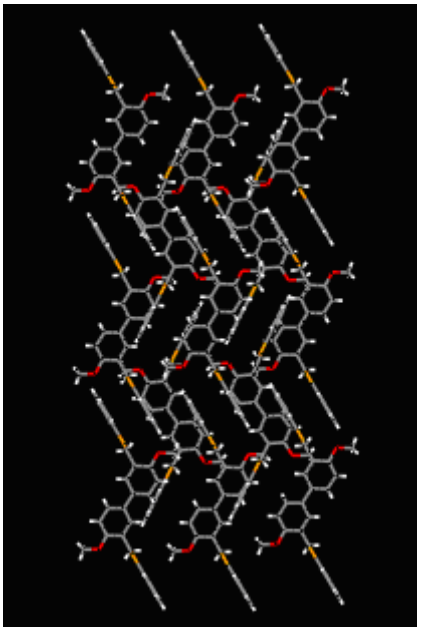


Table 1. Crystal data and structure refinement for 2C.

Identification code	f60904b
Empirical formula	C28 H26 O2 Se2
Formula weight	552.41
Temperature	273(2) K
Wavelength	0.71073 A
Crystal system, space group	Orthorhombic, Fdd2
Unit cell dimensions	a = 15.434(3) A alpha = 90 deg.
	b = 31.248(6) A beta = 90 deg.
	c = 9.855(2) A gamma = 90 deg.
Volume	4752.9(16) A^3
Z, Calculated density	8, 1.544 Mg/m^3

Absorption coefficient	3.135 mm^-1
F(000)	2224
Crystal size	0.15 x 0.15 x 0.10 mm
Theta range for data collection	2.54 to 28.25 deg.
Limiting indices	-20<=h<=19, -39<=k<=36, -13<=l<=8
Reflections collected / unique	5685 / 1462 [R(int) = 0.0409]
Completeness to theta = 28.25	94.0 %
Absorption correction	Semi-empirical from equivalents
Max. and min. transmission	0.7446 and 0.6507
Refinement method	Full-matrix least-squares on F^2
Data / restraints / parameters	1462 / 1 / 147
Goodness-of-fit on F^2	0.792
Final R indices [I>2sigma(I)]	R1 = 0.0393, wR2 = 0.1035
R indices (all data)	R1 = 0.0445, wR2 = 0.1093
Absolute structure parameter	0(10)
Largest diff. peak and hole	0.948 and -0.367 e.A^-3

Table 2. Atomic coordinates (x 10^4) and equivalent isotropic displacement parameters A^2 x 10^3) for **2C**. U(eq) is defined as one third of the trace of the orthogonalized Uij tensor.

	x	y	z	U(eq)
Se(1)	10127(1)	1523(1)	8527(1)	48(1)
O(1)	8278(2)	1318(1)	11036(6)	44(1)
C(1)	8713(3)	941(1)	10956(7)	34(1)
C(2)	8346(3)	542(2)	11184(7)	36(1)
C(3)	8844(3)	180(2)	11126(7)	37(1)
C(4)	9727(3)	198(1)	10835(6)	32(1)
C(5)	10079(3)	595(2)	10594(6)	34(1)
C(6)	9600(4)	968(2)	10647(5)	34(1)
C(7)	10034(4)	1394(2)	10441(6)	39(1)
C(8)	10718(3)	2058(2)	8604(8)	38(1)
C(9)	10964(5)	2248(2)	9802(7)	49(2)
C(10)	11394(6)	2641(2)	9787(8)	55(2)
C(11)	11580(4)	2841(2)	8555(11)	57(2)
C(12)	11336(6)	2647(3)	7377(9)	58(2)
C(13)	10903(5)	2256(2)	7378(7)	48(2)
C(14)	7402(4)	1304(2)	11524(8)	52(2)

Table 3. Bond lengths [A] and angles [deg] for **2C**.

Se(1)-C(8)	1.906(5)
Se(1)-C(7)	1.934(6)
O(1)-C(1)	1.357(5)
O(1)-C(14)	1.436(8)
C(1)-C(2)	1.389(7)
C(1)-C(6)	1.404(7)
C(2)-C(3)	1.368(7)
C(2)-H(2)	0.9300
C(3)-C(4)	1.395(7)
C(3)-H(3)	0.9300

C(4)-C(5)	1.374(7)
C(4)-C(4)#1	1.494(9)
C(5)-C(6)	1.382(7)
C(5)-H(5)	0.9300
C(6)-C(7)	1.506(7)
C(7)-H(7A)	0.9700
C(7)-H(7B)	0.9700
C(8)-C(9)	1.376(10)
C(8)-C(13)	1.388(10)
C(9)-C(10)	1.396(10)
C(9)-H(9)	0.9300
C(10)-C(11)	1.395(13)
C(10)-H(10)	0.9300
C(11)-C(12)	1.364(14)
C(11)-H(11)	0.9300
C(12)-C(13)	1.391(11)
C(12)-H(12)	0.9300
C(13)-H(13)	0.9300
C(14)-H(14A)	0.9600
C(14)-H(14B)	0.9600
C(14)-H(14C)	0.9600

C(8)-Se(1)-C(7)	100.3(3)
C(1)-O(1)-C(14)	117.4(4)
O(1)-C(1)-C(2)	124.6(4)
O(1)-C(1)-C(6)	116.3(4)
C(2)-C(1)-C(6)	119.1(4)
C(3)-C(2)-C(1)	120.4(5)
C(3)-C(2)-H(2)	119.8
C(1)-C(2)-H(2)	119.8
C(2)-C(3)-C(4)	121.7(4)
C(2)-C(3)-H(3)	119.2
C(4)-C(3)-H(3)	119.2
C(5)-C(4)-C(3)	117.2(4)
C(5)-C(4)-C(4)#1	121.6(5)
C(3)-C(4)-C(4)#1	121.2(5)
C(4)-C(5)-C(6)	123.0(5)
C(4)-C(5)-H(5)	118.5
C(6)-C(5)-H(5)	118.5
C(5)-C(6)-C(1)	118.6(4)
C(5)-C(6)-C(7)	120.2(5)
C(1)-C(6)-C(7)	121.1(4)
C(6)-C(7)-Se(1)	110.3(4)
C(6)-C(7)-H(7A)	109.6
Se(1)-C(7)-H(7A)	109.6
C(6)-C(7)-H(7B)	109.6
Se(1)-C(7)-H(7B)	109.6
H(7A)-C(7)-H(7B)	108.1
C(9)-C(8)-C(13)	119.8(5)
C(9)-C(8)-Se(1)	123.1(5)
C(13)-C(8)-Se(1)	117.1(5)
C(8)-C(9)-C(10)	120.2(7)

C(8)-C(9)-H(9)	119.9
C(10)-C(9)-H(9)	119.9
C(11)-C(10)-C(9)	120.1(7)
C(11)-C(10)-H(10)	120.0
C(9)-C(10)-H(10)	120.0
C(12)-C(11)-C(10)	119.0(5)
C(12)-C(11)-H(11)	120.5
C(10)-C(11)-H(11)	120.5
C(11)-C(12)-C(13)	121.5(7)
C(11)-C(12)-H(12)	119.2
C(13)-C(12)-H(12)	119.2
C(8)-C(13)-C(12)	119.4(7)
C(8)-C(13)-H(13)	120.3
C(12)-C(13)-H(13)	120.3
O(1)-C(14)-H(14A)	109.5
O(1)-C(14)-H(14B)	109.5
H(14A)-C(14)-H(14B)	109.5
O(1)-C(14)-H(14C)	109.5
H(14A)-C(14)-H(14C)	109.5
H(14B)-C(14)-H(14C)	109.5

Symmetry transformations used to generate equivalent atoms: #1 -x+2,-y,z

Table 4. Anisotropic displacement parameters (A^2 x 10^3) for **2C.** The anisotropic displacement factor exponent takes the form: -2 pi^2 [h^2 a*^2 U11 + ... + 2 h k a* b* U12]

	U11	U22	U33	U23	U13	U12
Se(1)	61(1)	42(1)	40(1)	-1(1)	-2(1)	-13(1)
O(1)	42(2)	32(2)	59(2)	-4(2)	-2(2)	7(1)
C(1)	36(2)	31(2)	34(2)	-2(2)	-5(3)	3(2)
C(2)	31(2)	39(2)	40(3)	0(2)	1(2)	-3(2)
C(3)	38(2)	30(2)	43(3)	1(3)	-1(3)	-6(2)
C(4)	31(2)	31(2)	35(3)	-1(2)	-2(2)	-1(2)
C(5)	29(2)	32(2)	39(3)	-2(2)	3(2)	-4(2)
C(6)	39(2)	28(2)	36(3)	-1(2)	-4(2)	-4(2)
C(7)	46(3)	31(2)	40(3)	2(2)	-1(2)	-5(2)
C(8)	36(2)	35(2)	43(3)	2(3)	6(3)	3(2)
C(9)	59(4)	45(4)	43(3)	7(3)	-4(3)	-13(3)
C(10)	64(5)	48(4)	52(4)	3(3)	-8(3)	-19(3)
C(11)	52(3)	40(3)	80(5)	4(4)	5(4)	-3(2)
C(12)	59(4)	53(4)	60(4)	24(3)	13(4)	8(3)
C(13)	51(4)	54(4)	40(3)	5(3)	4(3)	8(3)
C(14)	41(3)	52(3)	63(4)	-8(3)	-1(3)	12(3)

Table 5. Hydrogen coordinates (x 10^4) and isotropic displacement parameters (A^2 x 10^3) for **2C**.

	x	y	z	U(eq)
H(2)	7758	520	11378	44
H(3)	8587	-84	11285	44
H(5)	10665	613	10387	40
H(7A)	10608	1389	10843	47
H(7B)	9701	1616	10890	47
H(9)	10843	2115	10624	59
H(10)	11556	2770	10599	66
H(11)	11866	3103	8538	69
H(12)	11462	2778	6553	69
H(13)	10739	2129	6564	58
H(14A)	7060	1125	10938	78
H(14B)	7165	1588	11532	78
H(14C)	7394	1189	12427	78