Supplementary Material (ESI) for Chemical Communications

Electropolymerization of Intercalator-Grafted Conducting Polymer for Direct and Amplified DNA Detection

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Supporting Information

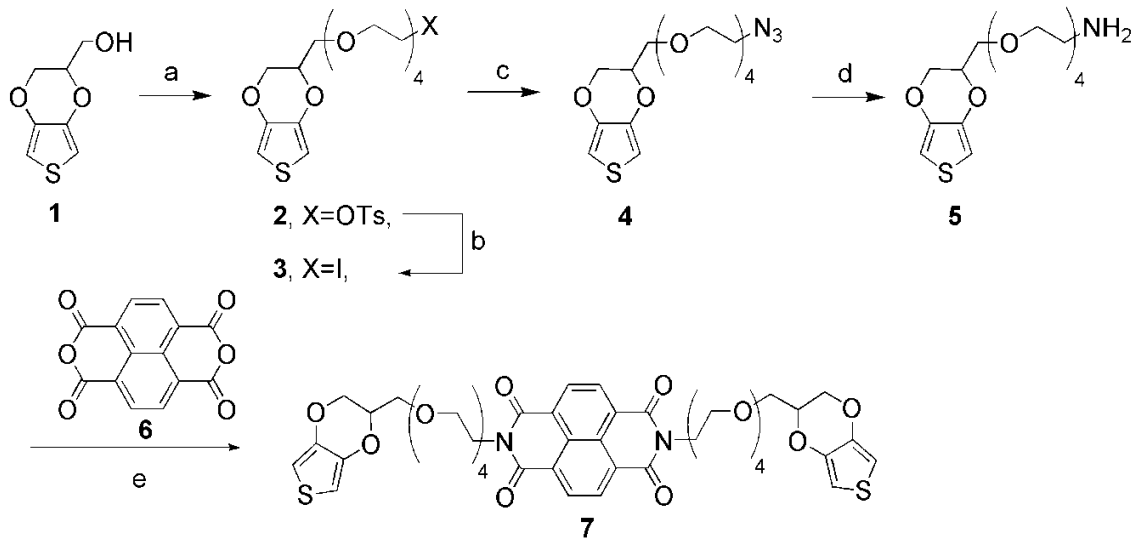
General Experimental

Hydroxymethyl EDOT was synthesized according to a previously described procedure.[S1] All chemicals were of reagent grade and used as received. Anhydrous solvents were purchased from Sigma-Aldrich in a sure-seal bottle, and introduced in the reaction flask under Ar using standard vacuum/inert gas manifold techniques. All other solvents were purchased from J. T. Baker (Phillipsburg, NJ). All reagents were purchased from commercial sources, and were used without further purification, unless otherwise indicated. Deuterated solvents were purchased from Sigma-Aldrich or Cambridge Isotope Laboratories, Inc. ^{1}H and ^{13}C NMR data were acquired at 25°C with a Bruker AV 400 spectrometer. Flash chromatography was performed on CombiFlash Companion or Rx16 on normal phase Silicagel cartridges. MS was carried out on a Finnigan/MAT LCQ Mass Spectrometer (ThermoFinnigan, San Jose, CA) fitted with an ESI probe. UV-Vis spectrophotometry was performed on an Agilent 8453 diode array spectrophotometer. Peptide nucleic acid (PNA) capture probe was custom synthesized by Applied Biosystems (Foster City, CA), while the DNA oligonucleotides were custom-made by 1st Base Pte Ltd (Singapore). The base sequences were N-TTTGAGTCTGTTGCTTGG (linker) – Cys (PNA capture probe), 5'-CCAAGCAACAGACTCAAA (complementary DNA target) and 5'-GGTTCGTTGTCTGAGTTT (non-complementary DNA target). Electrochemical study of EDOT intercalators was performed with an Autolab PGSTAT 32 potentiostat (Metrohm) in a glovebox from Innovative Technologies (Newburyport, MA). The one-chamber, three-electrode cell was made up of a quasi-internal Ag wire reference electrode (CH Instruments, Inc.) submerged in 0.01 M of $AgNO_3$/0.1 M of $(n\text{Bu})_4NPF_6$ in anhydrous CH_3CN, a platinum button working electrode, and a platinum coil counter electrode. Electrochemical DNA detection was carried out using a CH Instruments model 760C electrochemical workstation (CH Instruments, Austin, TX). A conventional three-electrode system, consisting of a 3.0-mm diameter gold working electrode (CH Instruments), a nonleak miniature Ag/AgCl reference electrode (Cypress Systems, Lawrence, KS), and a platinum wire counter electrode, was used in all electrochemical measurements. PBS buffer (15 M of NaCl, 20 mM of phosphate buffer; pH 7.4) was used for immobilization of PNA capture probes, while TE buffer (Tris-HCl, 1.0 mM of EDTA, pH 8.0) containing 0.1 M of NaCl and 0.01% of Triton X was used as hybridization buffer and for post-hybridization washing. The intercalator was incubated in TE buffer; while a NaCl-saturated TE

buffer containing 10% of ethanol was used as the final washing solution before analysis. Aqueous $LiClO_4$ solution (0.1 M) was used as electrolyte in electrochemical detection procedure.

Synthetic Procedures

Synthesis of EDOT-ND-EDOT



a) NaH, tetraethyleneglycol ditosylate, 18-crown-6, THF, reflux, 18 h, 20%. b) NaI, acetone, reflux, 18 h, 44%. c) NaN_3, DMF/H_2O, 100°C, 18 h, 65%. d) 1. PPh_3, THF, 50°C, 1 h. 2. NaOH, THF/H_2O, 50°C, 2 h, 86%. e) $Zn(OAc)_2$, pyridine, reflux, 18 h, 30%.

11-((2,3-dihydrothieno[3,4-b][1,4]dioxin-3-yl)methyl)-3,6,9-trioxaundecyl tosylate (EDOT-EG4-OTs, 2). A solution of EDOT-OH (**1**; 1.72 g, 10.0 mmol) and 18-crown-6 (132 mg, 0.500 mmol) in anhydrous THF (10 mL) was added dropwise to NaH (95%, 1.26 g, 50.0 mmol) suspended in anhydrous THF (150 mL) at 0°C. The mixture was then added to tetraethyleneglycol ditosylate (1.01 g, 20.0 mmole) and was refluxed overnight under N_2. After quenching with water, THF was removed *in vacuo* and extracted three times with CH_2Cl_2. The combined organic phase was dried with $MgSO_4$, and purified by flash chromatography (dichloromethane/ethyl acetate = 9:1). EDOT-EG4-OTs (**2**; 1.00 g, 20%) was obtained as light yellow liquid after drying under vacuum. ^{1}H NMR (400 MHz, $CDCl_3$): δ 7.80 (d, 1H, *J* = 1.6 Hz), 7.78 (d, 1H, *J* = 1.2 Hz), 6.32 (d, 1H, *J* = 3.6 Hz), 6.31 (d, 1H, *J* = 3.6 Hz), 4.35–4.29 (m, 1H), 4.24 (dd, 1H, *J* = 11.6, 2.0 Hz), 4.15 (t, 2H, *J* = 4.8 Hz), 4.11 (dd, 1H, *J* = 14.4, 7.2 Hz), 4.05 (dd, 1H, *J* = 11.6, 7.6 Hz), 3.76 (dd, 1H, *J* = 10.4, 4.8 Hz), 3.71–3.53 (m, 14H), 2.44 (s, 3H). ^{13}C

NMR (100 MH, $CDCl_3$) 145.0, 145.0, 141.7, 141.6, 132.9, 130.0, 128.1, 99.8, 99.7, 72.7, 71.3, 70.8, 70.8, 70.7, 70.6, 70.7, 70.7 , 70.6, 69.7, 69.4, 68.8, 66.2, 21.8. HR-MS (FAB): calcd. for $C_{22}H_{30}O_9S_2+H^+$ [M+H$^+$] 503.1404; found 503.1394.

2-((11-iodo-3,6,9-trioxaundecyloxy)methyl)-2,3-dihydrothieno[3,4-b][1,4]dioxine (EDOT-EG4-I, 3). A solution of **2** (1.00 g, 1.99 mmol) and sodium iodide (1.49 g, 9.95 mmol) was refluxed in acetone (20 mL) for 18 h. The reaction mixture was then filtered, and the volatiles was removed *in vacuo*, dissolved in CH_2Cl_2, and washed with saturated $Na_2S_2O_{5(aq)}$. After purification by flash chromatography (dichloromethane/ethyl acetate = 19:1), EDOT-EG4-I (**3**; 400 mg, 44%) was obtained as light yellow liquid after drying under vacuum. ^{1}H NMR (400 MHz, $CDCl_3$): δ 6.33 (d, 1H, *J* = 3.6 Hz), 6.32 (d, 1H, *J* = 3.6 Hz), 4.36–4.29 (m, 1H), 4.25 (dd, 1H, *J* = 11.6, 2.8 Hz), 4.06 (dd, 1H, *J* = 11.6, 7.2 Hz), 3.77 (dd, 1H, *J* = 9.6, 4.8 Hz), 3.75 (t, 2H, *J* = 5.2 Hz), 3.71–3.63 (m, 13H), 3.26 (t, 2H, *J* = 7.2 Hz). ^{13}C NMR (100 MH, $CDCl_3$) 141.7, 141.6, 99.9, 99.8, 72.8, 72.1, 71.3, 70.8, 70.8, 70.7, 70.7, 69.8, 66.3, 53.7. HR-MS (FAB): calcd. for $C_{15}H_{23}IO_6S+H^+$ [M+H$^+$] 459.0333; found 459.0317.

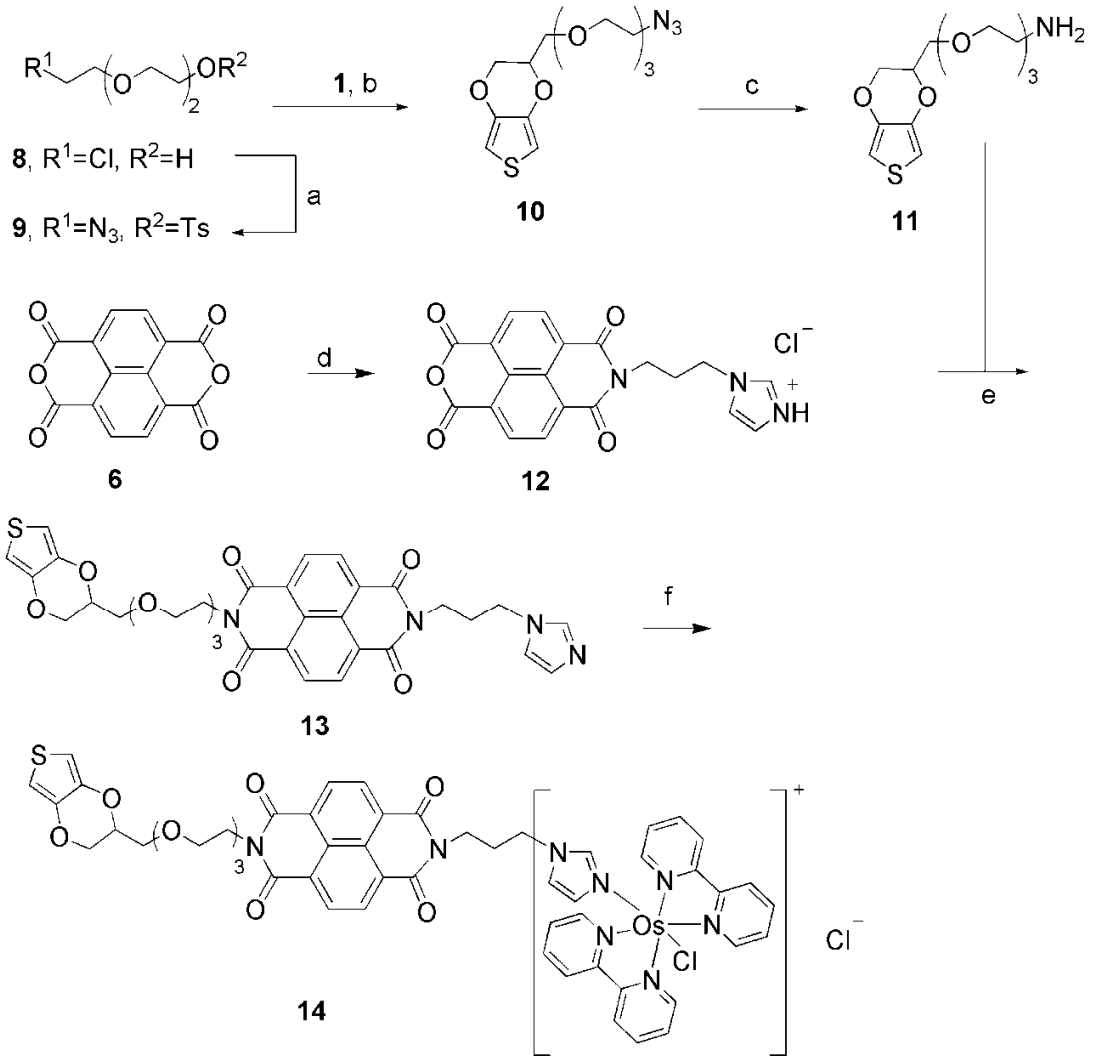
2-((11-azido-3,6,9-trioxaundecyloxy)methyl)-2,3-dihydrothieno[3,4-b][1,4]dioxine (EDOT-EG4-N_3, 4) A solution of **3** (400 mg, 0.873 mmol) in DMF (5 mL) and a solution of sodium azide (227 mg 3.49 mmol) in water (5 mL) were mixed together and refluxed for 18 h. DMF was removed by washing with saturated $NH_4Cl_{(aq)}$. The reaction mixture was dissolved in CH_2Cl_2, washed with water, and dried with $MgSO_4$. The crude product was purified by flash chromatography (dichloromethane/ethyl acetate = 19:1) to yield a viscous colorless liquid (212 mg, 0.568 mmol, 65%). ^{1}H NMR (400 MHz, $CDCl_3$): δ 6.33 (d, 1H, *J* = 4 Hz), 6.32 (d, 1H, *J* = 3.6 Hz), 4.35–4.29 (m, 1H), 4.25 (dd, 1H, *J* = 11.6, 2.4 Hz), 4.06 (dd, 1H, *J* = 11.6, 7.2 Hz), 3.76 (dd, 2H, *J* = 10.8, 5.2 Hz), 3.71–3.52 (m, 15H), 3.39 (t, 2H, *J* = 5.2 Hz). ^{13}C NMR (100 MHz, $CDCl_3$): δ 141.6, 141.5, 99.7, 99.6, 77.3, 72.6, 71.2, 70.7, 70.7, 70.6, 70.5, 70.1, 69.6, 66.1, 50.7. HR-MS (FAB): calcd. for $C_{15}H_{23}N_3O_6S+H^+$ 374.1386 [M+H$^+$]; found 374.1379.

2-((11-amino-3,6,9-trioxaundecyloxy)methyl)-2,3-dihydrothieno[3,4-b][1,4]dioxine EDOT-EG4-NH_2. A solution of **4** (100 mg, 0.268 mmol) in THF (3 mL) was mixed with triphenylphosphine (77.3 mg, 0.295 mmol), and was heated to 50°C for 1 h. 3 mL of $NaOH_{(aq)}$ (2

M) was subsequently added, and the reaction was stirred for another 2 h. THF was removed by rotary evaporator, and the aqueous reaction mixture was acidified to pH < 3. The aqueous phase was washed with CH_2Cl_2. NaOH was then added, and the resulting solution (pH > 10) was extracted with CH_2Cl_2. The organic layer was dried with Na_2SO_4, and the solvent was removed *in vacuo* to give a viscous yellow liquid (80.0 mg, 86%). ^{1}H NMR (400 MHz, $CDCl_3$): δ 6.29 (d, 1H, *J* = 3.6 Hz), 6.28 (d, 1H, *J* = 3.2 Hz), 4.32–4.25 (m, 1H), 4.21 (dd, 1H, *J* = 11.6, 2 Hz), 4.02 (dd, 1H, *J* = 11.6, 7.6 Hz), 3.72 (dd, 2H, *J* = 10.4, 4.8 Hz), 3.67–3.57 (m, 15H), 3.47 (t, 2H, *J* = 5.2 Hz). ^{13}C NMR (100 MHz, $CDCl_3$): δ 141.5, 141.4, 99.7, 99.6, 72.9, 72.6, 71.1, 70.6, 70.5, 70.5, 70.2, 69.6, 66.1, 53.5, 41.5. HR-MS (FAB): calcd. for $C_{15}H_{25}NO_6S+H^+$ 348.1481 [M+H$^+$]; found 348.1478.

***N,N'*-bis[(11-(2,3-dihydrothieno[3,4-b][1,4]dioxin-3-yl)methyl)-3,6,9-trioxaundecyl]-1,4,5,8-naphthalenetetracarboxydiimide (Bis-EDOT-ND, 7).** Dianhydride **6** (35.1 mg, 0.131 mmol), the amine **5** (100.0 mg, 0.288 mmol), and zinc acetate (20.2 mg, 0.092 mmol) were refluxed in pyridine (10 mL) over 18 h. The reaction mixture was filtered through a short column of silica gel with CH_2Cl_2 as eluent. The organic solution was then washed with HCl (1 N) and deionized water, and dried with $MgSO_4$; the solvent was removed by a rotary evaporator. The diimide **7** was obtained as an orange solid (11.0 mg, 30%) after flash chromatography (dichloromethane/ethyl acetate = 2:1). ^{1}H NMR (400 MHz, $CDCl_3$): δ 8.75 (s, 4H), 6.31 (d, 2H, *J* = 3.6 Hz), 6.29 (d, 2H, *J* = 3.6 Hz), 4.46 (t, 4H, *J* = 6 Hz), 4.34–4.27 (m, 2H), 4.23 (dd, 2H, *J* = 11.6, 2.4 Hz), 4.04 (dd, 2H, *J* = 11.6, 4.8 Hz), 3.84 (t, 4H, *J* = 5.6 Hz), 3.75 (dd, 2H, *J* = 10.4, 4.8 Hz), 3.72–3.64 (m, 10H), 3.64–3.56 (m, 16H). ^{13}C NMR (100 MHz, $CDCl_3$): δ 162.9, 141.5, 141.5, 131.0, 126.7, 126.6, 100.0, 99.7 99.6, 77.9, 72.6, 71.2, 70.6, 70.6, 70.5, 70.1, 69.6, 67.8, 66.1, 39.6. HR-MS (FAB): calcd. for $C_{44}H_{50}N_2O_{16}S_2+H^+$ 927.2680 [M+H$^+$]; found 927.2698.

Synthesis of EDOT-ND-Os



a) 1. NaN_3, cat. NaI, H_2O, 80°C, 18 h. 2. TsCl, cat. DMAP, Et_3N, CH_2Cl_2, RT, 18 h, 94%. b) **1**, NaH, cat. NaI, DMF, RT, 18 h, 89%. c) 1. PPh_3, THF, 50°C, 1 h. 2. NaOH, THF/H_2O, 50°C, 2 h, 64%. d) 1. 1(3-aminopropyl)imidazole, DMA, 125°C, 18 h. 2. $SOCl_2$, DMF, 2 h, 55°C, 61%. e) **11**, $Zn(OAc)_2$, pyridine, 120°C, 18 h, 18%. f) $[Os(bpy)_2Cl_2]Cl{\cdot}2H_2O$, ethyleneglycol, 180°C, 6 h, 33%.

8-azido-3,6-dioxaoctyl tosylate (TsO-EG3-N_3, 9). Chloride **8** (1.46 mL, 1.69 g, 10 mmol) was added to a solution of NaN_3 (3.25 g, 50 mmol) and NaI (0.30 g, 2 mmol), and the mixture was heated at 80°C over 18 h. The solution was transferred to a separatory funnel and extracted with CH_2Cl_2 (5×). The combined organic layers were dried ($MgSO_4$), and the solution volume was reduced to ~ 20–30 mL. Tosyl chloride (2.10 g, 11 mmol) and 4-dimethylaminopyridine (DMAP; 122 mg, 1 mmol) were added, followed by the dropwise addition of Et_3N (1.6 mL, 1.21 g, 12

mmol). After 18 h, the solution was washed with 10% $H_2SO_{4(aq)}$, saturated with $NaHCO_{3(aq)}$, and dried ($MgSO_4$); the volatiles were removed in vacuum. The azide **9** (3.10 g, 94%) was obtained as a colorless liquid after column chromatography (CombiFlash 40 g cartridge, 0 to 60% gradient of ethyl acetate in hexane over 20 min). The ^{1}H and ^{13}C NMR data were in agreement with those previously reported.[S2]

2-((8-azido-3,6-trioxaoctyloxy)methyl)-2,3-dihydrothieno[3,4-b][1,4]dioxine (EDOT-EG3-N_3, 10). NaH (60% in mineral oil; 600 mg, 15 mmol) was added to a solution of EDOT-OH (**1**; 1.72 g, 10 mmol), NaI (0.375 g, 2.5 mmol) and dry DMF (10 mL) against a weak back-flow of Ar, and the mixture was stirred for 20 min. A solution of **9** (3.29 g) in DMF (10 mL) was added dropwise, and the mixture was stirred over 18 h. The mixture was partitioned between H_2O (300 mL) and diethyl ether (100 mL), and the organic layer was further washed with H_2O (5×). After drying ($MgSO_4$) and removal of the volatiles in vaccum, **10** (2.94 g, 89%) was obtained after column chromatography (CombiFlash 40 g cartridge, 30 to 70% gradient of ethyl acetate in hexane over 20 min). ^{1}H NMR (400 MHz, $CDCl_3$): δ 6.35 (d, 1H, *J* = 3.6 Hz), 6.34 (d, 1H, *J* = 3.2 Hz), 4.36–4.31 (m, 1H), 4.26 (dd, 1H, *J* = 10.8, 1.6 Hz), 4.07 (dd, 1H, *J* = 10.8, 7.6 Hz), 3.77 (dd, 2H, *J* = 10.8, 5.2 Hz), 3.70–3.67 (m, 10H), 3.39 (t, 2H, *J* = 4.8 Hz). ^{13}C NMR (100 MHz, $CDCl_3$): δ 141.6, 141.5, 99.7, 99.6, 72.6, 71.2, 70.7, 70.6, 70.1, 69.6, 66.1, 60.4, 50.1.

2-((8-amino-3,6-dioxaoctyloxy)methyl)-2,3-dihydrothieno[3,4-b][1,4]dioxine (EDOT-EG3-NH_2, 11). A solution of **10** (660 mg, 2.0 mmol) in THF (10 mL) and triphenylphosphine (577 mg, 2.2 mmol) was heated to 50°C for 1 h. $NaOH_{(aq)}$ (2 M; 10 mL) was added, and the reaction mixture was stirred for another 2 h. THF was removed by rotary evaporator, and the aqueous reaction mixture was acidified to pH < 3. The aqueous phase was extracted with CH_2Cl_2 (3×), and the organic layers were discarded. NaOH was then added to the aqueous layer, and the solution (pH > 10) was extracted with CH_2Cl_2 (3×). The organic layer was dried with Na_2SO_4, and the solvent was removed *in vacuo* to give a viscous yellow liquid (388 mg, 86%). ^{1}H NMR (400 MHz, $CDCl_3$): δ 6.29 (d, 1H, *J* = 3.6 Hz), 6.28 (d, 1H, *J* = 3.2 Hz), 4.32–4.25 (m, 1H), 4.21 (dd, 1H, *J* = 11.6, 2 Hz), 4.02 (dd, 1H, *J* = 11.6, 7.6 Hz), 3.72 (dd, 2H, *J* = 10.4, 4.8 Hz), 3.67–3.57 (m, 15H), 3.47 (t, 2H, *J* = 5.2 Hz). ^{13}C NMR (100 MHz, $CDCl_3$): δ 141.5, 141.4, 99.7, 99.6,

72.9, 72.6, 71.1, 70.6, 70.5, 70.5, 70.2, 69.6, 66.1, 53.5, 41.5. HR-MS (FAB): calcd. for $C_{13}H_{21}NO_5S+H^+$ 304.1213 [$M+H^+$]; found 304.1210.

***N*-[3-(imidazol-1-yl)propyl]-*N'*-[8-((2,3-dihydrothieno[3,4-b][1,4]dioxin-3-yl)methoxy)-3,6-dioxaoctyl]-1,4,5,8-naphthalenetetracarboxydiimide (EDOT-ND-Im, 13).** A mixture of **6** (804.6 mg, 3 mmol) and 1-(3-aminopropyl)imidazole (125.2 mg, 1 mmol) in dimethylacetate (30 mL) was heated at 125°C for 18 h. $CHCl_3$ (100 mL) was added upon cooling to room temperature. The precipitate was filtered, and the volatiles were removed in vacuum. Water (150 mL) was added and the resulting precipitate was washed with ethanol and ether. The crude product was heated with $SOCl_2$ (142.8 mg, 1.2 mmol) in DMF at 60°C for 2 h. The resulting precipitate was collected and washed with ether to produce **12** (251.2 mg, 0.61 mmol, 61%), which was used for the next step without further purification. A mixture of **11** (50 mg, 0.17 mmol), **12** (70 mg, 0.17 mmol) and zinc acetate (25.4 mg, 0.12 mmol) was heated in pyridine at 120°C for 15 h. After removal of pyridine by vacuum distillation, the reaction mixture was partitioned between CH_2Cl_2 and Na. The combined organic layer was dried with $MgSO_4$, and subjected to further purification by flash chromatography (dichloromethane/ethyl acetate = 1/9). The product was obtained as yellow gel (18.5 mg, 0.03 mmol, 17.6% yield). ^{1}H NMR (400 MHz, $CDCl_3$): δ 8.74 (t, 4H, *J* = 8 Hz), 7.56 (m, 1H), 7.02 (m, 2H), 6.32 (d, 1H, *J* = 3.6 Hz), 6.31 (d, 1 H, *J* = 3.6 Hz), 4.48 (t, 2H, *J* = 6 Hz), 4.32–4.25 (m, 1H), 4.23 (dd, 1H, *J* = 11.6, 2 Hz), 4.12 (t, 2H, *J* = 7.6 Hz), 4.05 (dd, 1H, *J* = 11.6, 7.2 Hz), 3.87 (dd, 1H, *J* = 10.4, 6 Hz), 3.71 (t, 2H, *J* = 5.6 Hz), 3.66–3.60 (m, 8 H), 2.32 (q, 2H, *J* = 5.6 Hz), ^{13}C NMR (400 MHz, $CDCl_3$): δ 163.1, 163.0, 141.7, 141.6, 137.4, 131.4, 131.2, 129.9, 127.0, 126.9, 126.9, 126.5, 118.8, 100.2, 99.8, 72.8, 71.4, 70.9, 70.7, 70.3, 69.8, 68.0, 66.3, 45.1, 39.8, 38.5, 29.5. HR-MS (FAB): calcd. for $C_{33}H_{32}N_4O_9S+H^+$ 661.1963 [$M+H^+$]; found 661.1972.

***N*-[3-(imidazol-1-yl)propyl]-*N'*-[8-((2,3-dihydrothieno[3,4-b][1,4]dioxin-3-yl)methoxy)-3,6-dioxaoctyl]-1,4,5,8-naphthalenetetracarboxydiimide complex with $Os(bpy)_2Cl_2$ (EDOT-NTCDI-Os, 14).** $[Os(bpy)_2Cl_2]Cl\cdot 2H_2O$[S3] (18.9 mg, 0.029 mmol) was added to a solution of **13** (18.5 mg, 0.030 mmol) in ethylene glycol, and the mixture was stirred at 180°C over 6 h. The progress of the reaction was monitored by cyclic voltammetry. Upon completion, ethylene glycol was removed, and the solid was extracted with $CHCl_3$ (3×). The combined $CHCl_3$ layers

were washed repeatedly with water. The product was obtained as dark purple paste (14.2 mg, 33% yield). HR-MS (ESI): calcd. for $C_{53}H_{48}ClN_8O_9OsS^+$ 1199.2563 [M^+]; found 1199.2571.

Electrochemical Experiments and DNA Detection

Immobilization of PNA capture probe (CP) on gold electrode. The gold button electrode was first mechanically polished with 0.5-μm alumina slurry, followed by ultrasonication in IPA and water. Electrochemical cleaning was subsequently done by repeated potential scanning at -0.3 to 1.5 V (vs. Ag/AgCl) in 0.1 M of H_2SO_4 solution. Upon washing in water and drying with a stream of nitrogen, the electrode was ready for use. A monolayer of CP was adsorbed by immersing the gold electrode in a 1-μM solution of CP in PBS for 12 h. After adsorption, the electrode was copiously rinsed with PBS, and blown dry with a stream of nitrogen. To minimize non-specific uptake of target DNA and intercalator, and improve the quality and stability of the CP monolayer, the CP-coated gold electrode was immersed in an ethanolic solution of 1 mM of 11-mercaptoundecanol (MUD) for 3 h. Unreacted MUD was rinsed with ethanol and then with water. Upon blow drying with nitrogen, the electrode was ready for the next step.

Hybridization and detection. The target DNA was hybridized in a moisture-saturated chamber maintained at 37°C. A 2.5-μl aliquot of hybridization solution containing the target DNA was uniformly spread onto the CP-coated electrode, and left to hybridize for 4 h. To remove non-specifically bounded DNA from the electrode surface, a series of high- and low-stringency washes was carried out. The electrode was first washed in a stirred hybridization buffer (blank, with 0.01% of triton-X) at 37°C for 5 min, followed by immersion in blank TE buffer at room temperature for 1 min, and a brief wash in water. A 3.0-μl aliquot of 100 μM of EDOT-ND–Os in TE buffer was then added to the electrode surface, allowing it to incubate at room temperature for 15 min. After thorough rinsing using the final washing solution, the electrode was ready for the electrochemical analysis (Scheme S1). First, the redox reaction of Os^{+2}/Os^{+3} was observed through square-wave voltammetry in 0.1 M solution of $LiClO_{4\ (aq)}$ (see Figure 2A). Next, the electrode was subjected to five cycles of potential scan at -0.2 to 1.0 V (vs. Ag/AgCl) to form oligoEDOTs. These serve as ‘seeds’ for subsequent polymerization. In the final step, the electrode was immersed in 0.1 M of $LiClO_{4\ (aq)}$ solution containing 5.0 mM of EDOT-OH. A

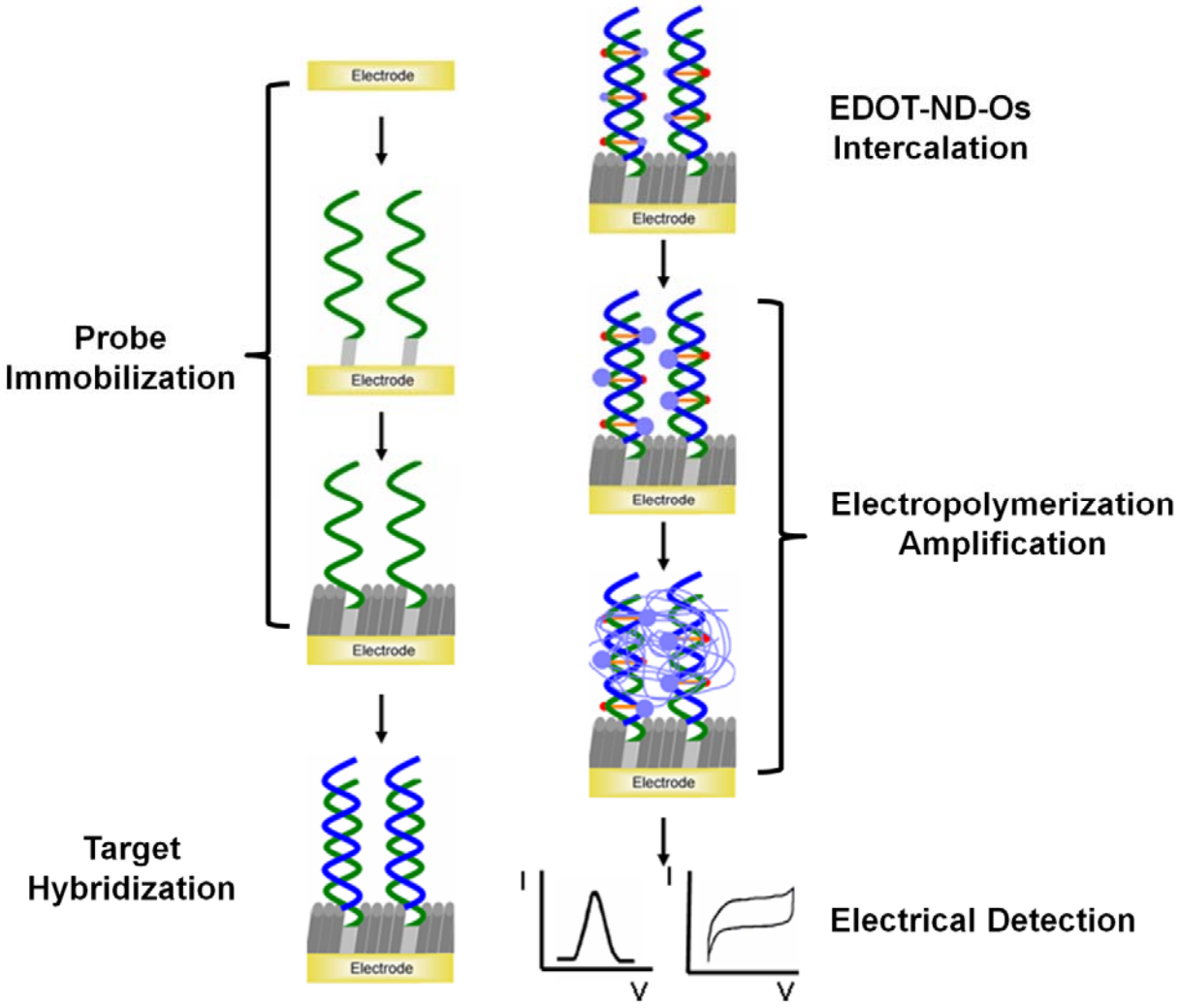
constant potential was applied at 0.9 V (vs. Ag/AgCl) for 120 sec to allow polymerization of the EDOT-OH monomers. After a brief washing in water, the electrode was analyzed in a blank electrolyte solution to quantify the copolymer film that has been formed (see Figure 2C). The original detection data was described in Figure S1.

References

[S1] (a) A. Lima, P. Schottland, S. Sadki, C. Chevrot, *Synthetic Metals* **1998**, *93*, 33. (b) S. Akoudad, J. Roncali, *Electrochemistry Communications* **2000**, *2*, 72.

[S2] S. J. Meunier, Q. Wu, S.-N. Wang, R. Roy, *Can. J. Chem.* **1997**, *75*, 1472.

[S3] P. A. Lay, A. M. Sargeson, H. Taube. *Inorg. Synth.* **1986**, *24*, 291.



Scheme S1. Representation of electrochemical DNA detection using EDOT-grafted intercalator.

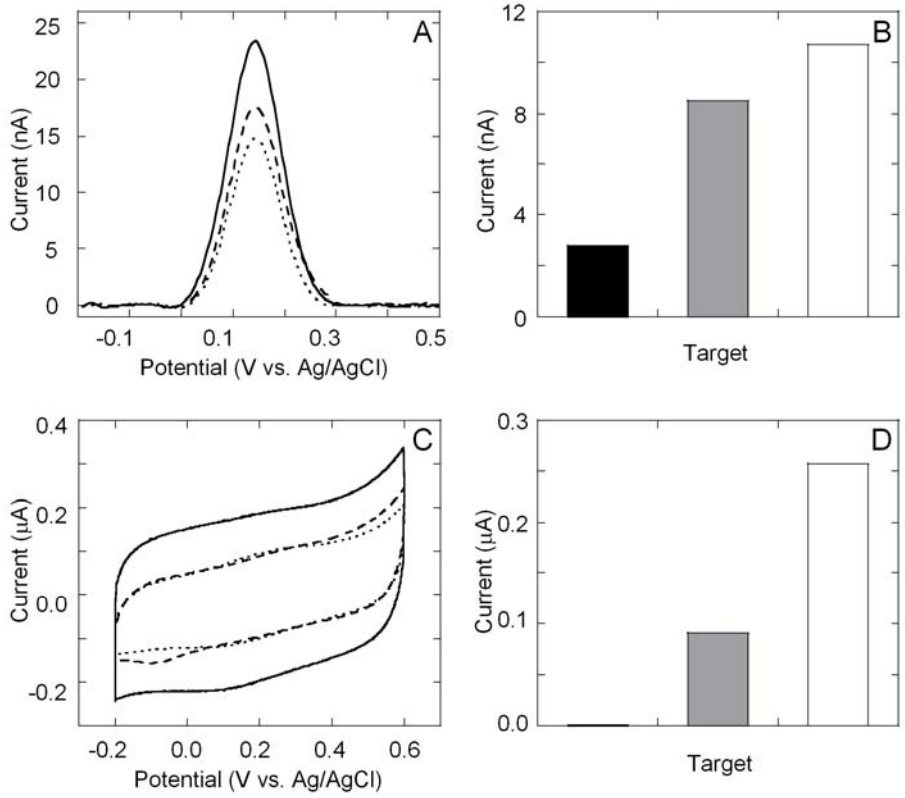


Figure S1. (A) Square-wave voltammograms of EDOT-ND-Os bound to DNA capture probe hybridized with 20 pM of complementary target (—), 100 pM of non-complementary target (---), and no target (···). (B) Peak current from biosensor electrodes hybridizing with (□) 100 pM of complementary target, (■) 20 pM of complementary target, and (■) 100 pM of non-complementary target from (A) at 0.14 V after signal subtraction of blank experiment (no target). (C) Cyclic voltammograms of PEDOTs formed on assembled biosensor electrodes as described in (A) after seed-mediated electropolymerization of 5 mM of EDOT–OH. (D) Voltammetric current from biosensor electrodes hybridizing with (□) 100 pM of complementary target, (■) 20 pM of complementary target, and (■) 100 pM of non-complementary target from (C) at 0.3 V (oxidation) after signal subtraction of blank experiment (no target). The voltammograms were measured in aqueous solution containing 0.1 M of $LiClO_4$ as supporting electrolyte.

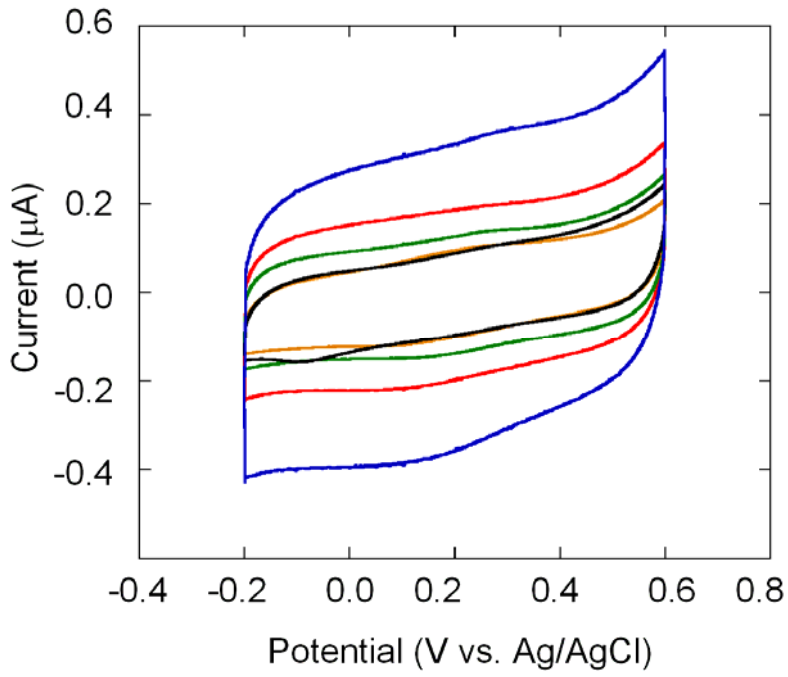


Figure S2. Cyclic voltammograms of poly(EDOT)s formed after seed-mediated electropolymerization of 5 mM of EDOT-OH on biosensors with no target (orange), 100 pM noncomplementary target (black), 20 pM two-base mismatched target (green), 20 pM complementary target (red), and 100 pM complementary target (blue). Voltammograms were recorded in an aqueous solution of 0.1 M $LiClO_4$ at a scan rate of 100 mV/s.

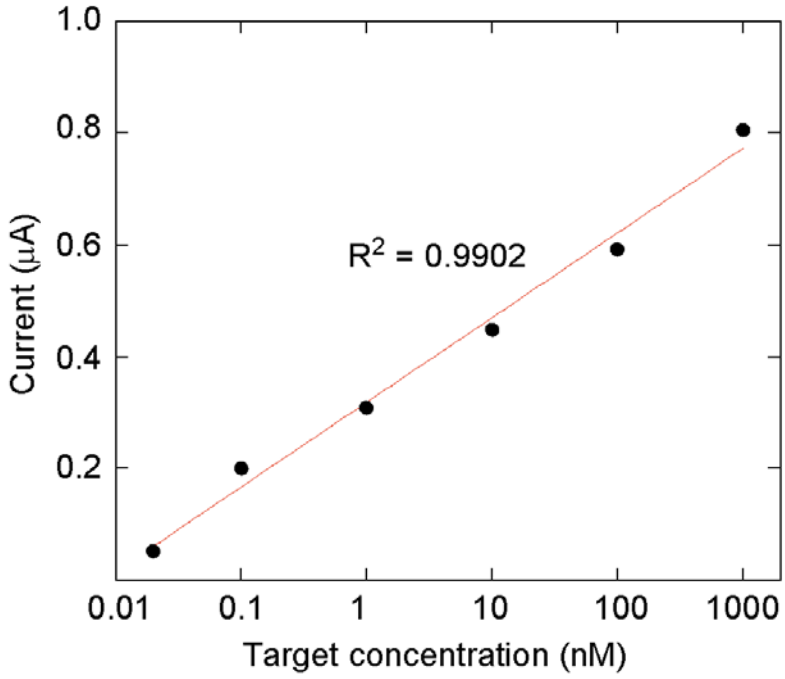


Figure S3. Calibration plot based on the voltammetric current from biosensor electrodes with complementary target at various concentrations. Values taken at 0.3 V after substraction of the signal from blank experiment (without target).

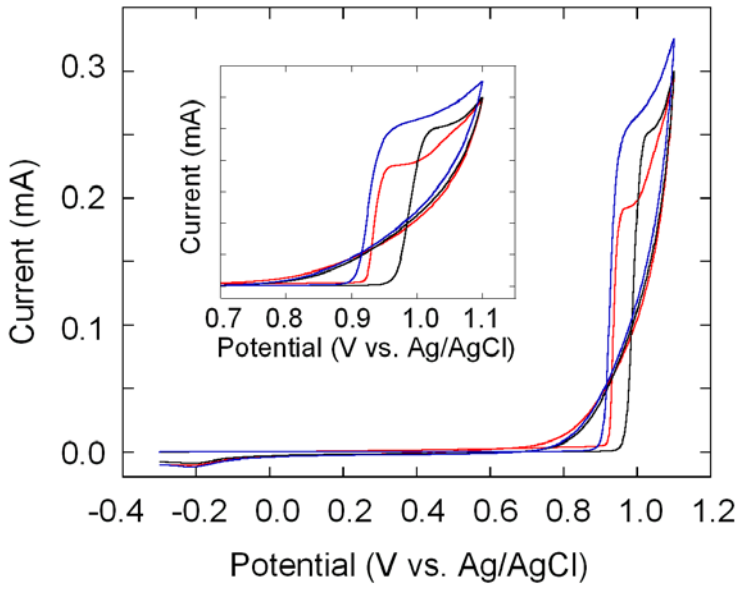
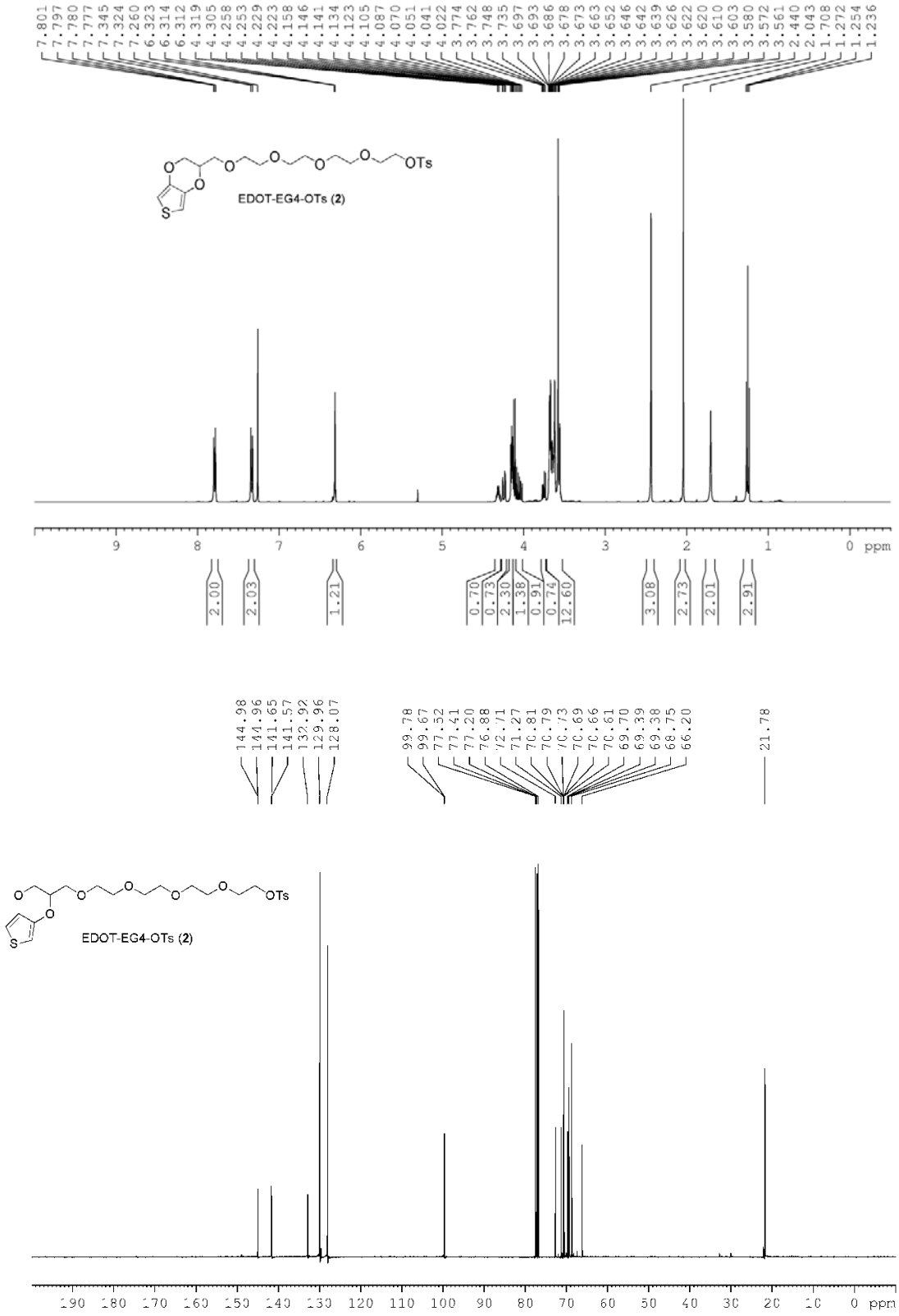
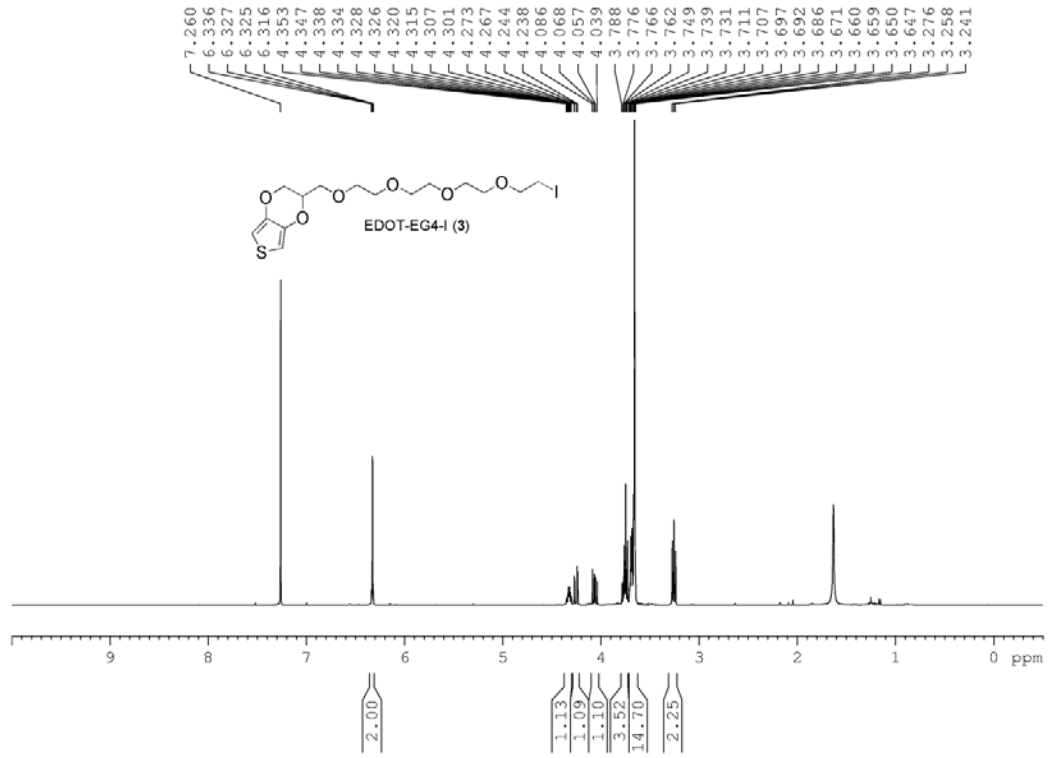
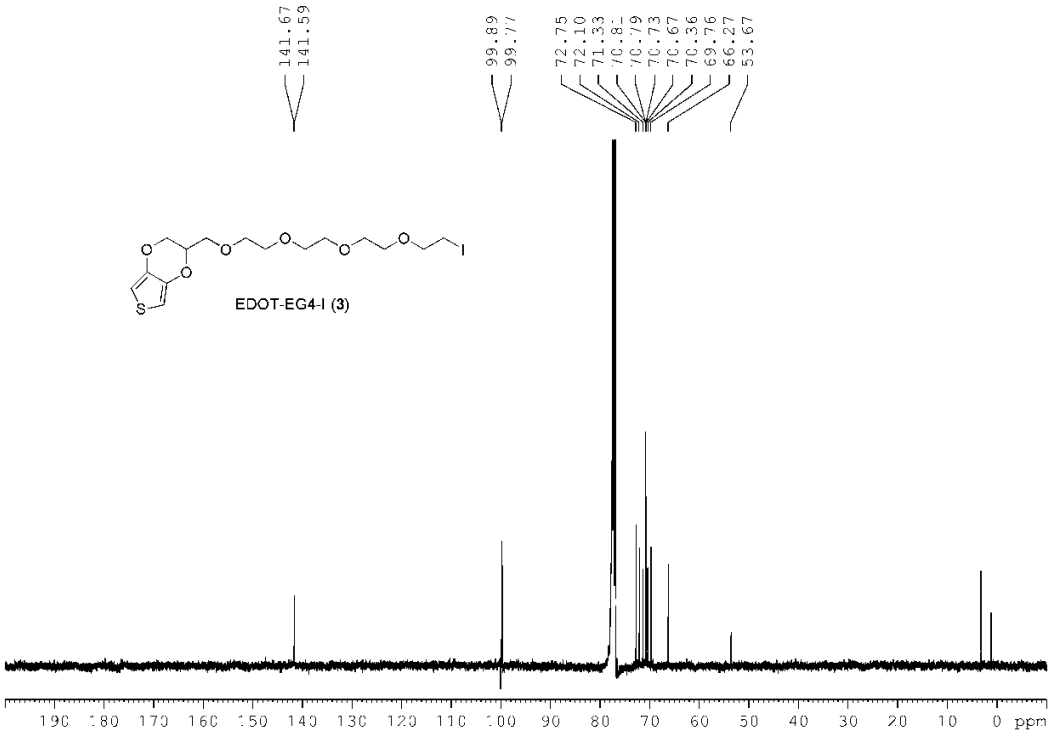


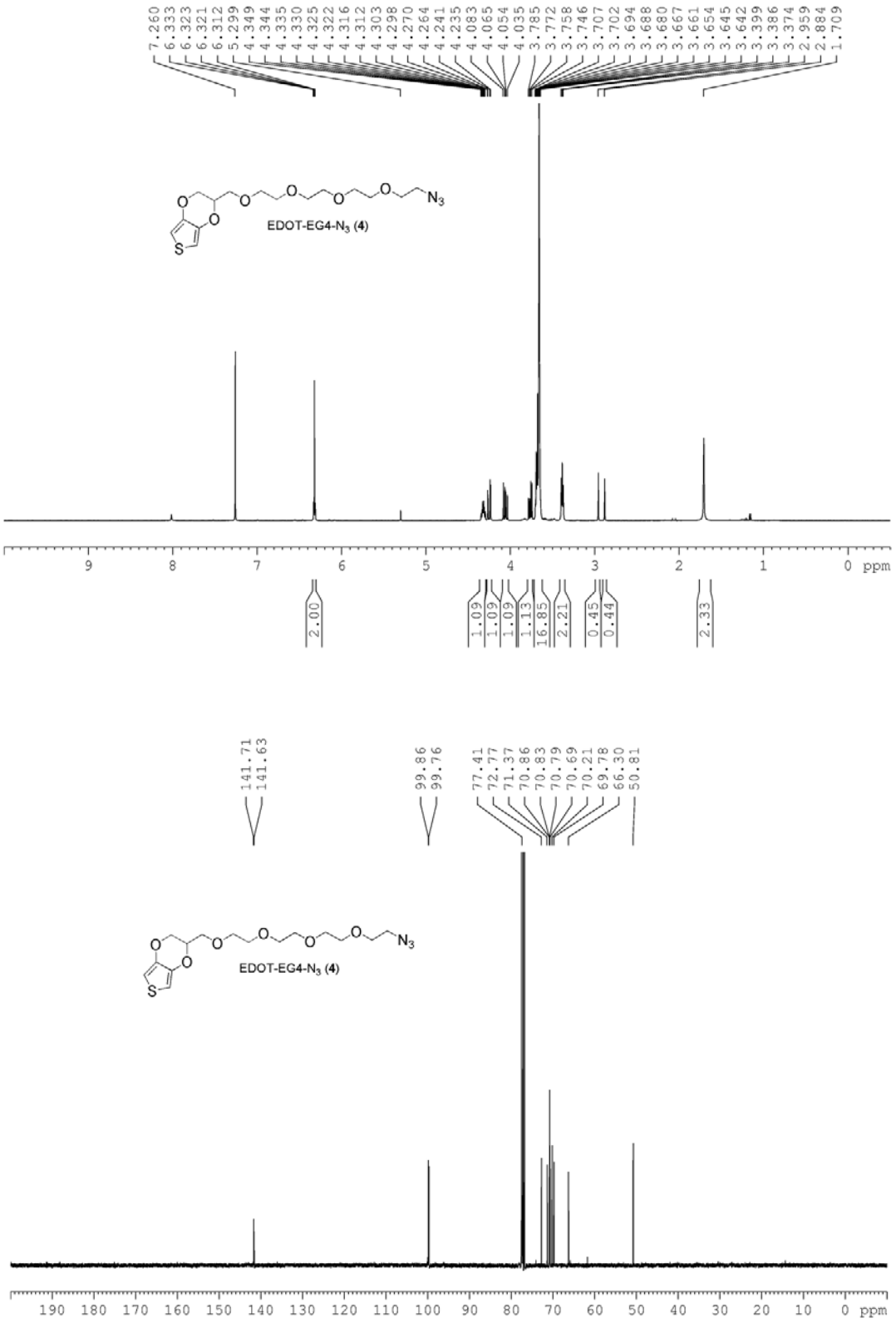
Figure S4: Cyclic voltammograms of EDOT-OH oxidation on Au electrode with various modifications: SAM of mercaptoundecanol (—), mixed monolayer of capture probe/mercaptoundecanol and target DNA (—), and complete biosensor with mixed monolayer of capture probe/mercaptoundecanol, target DNA, and reporter molecule EDOT-ND-Os (—).

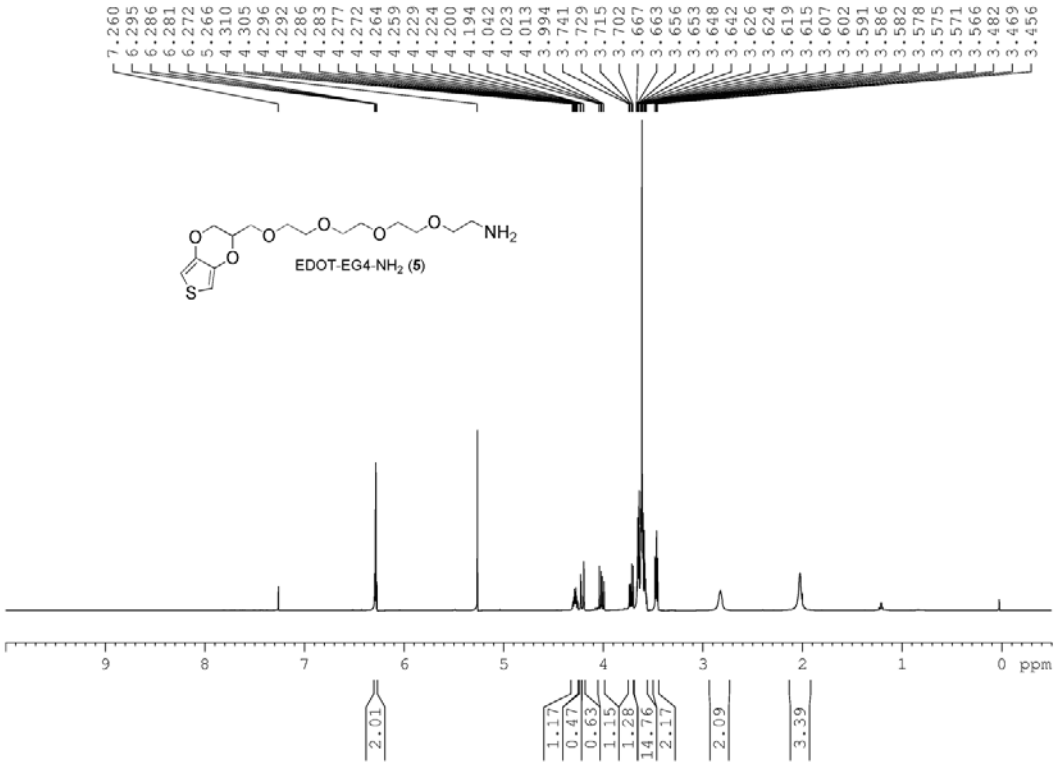
NMR Spectra

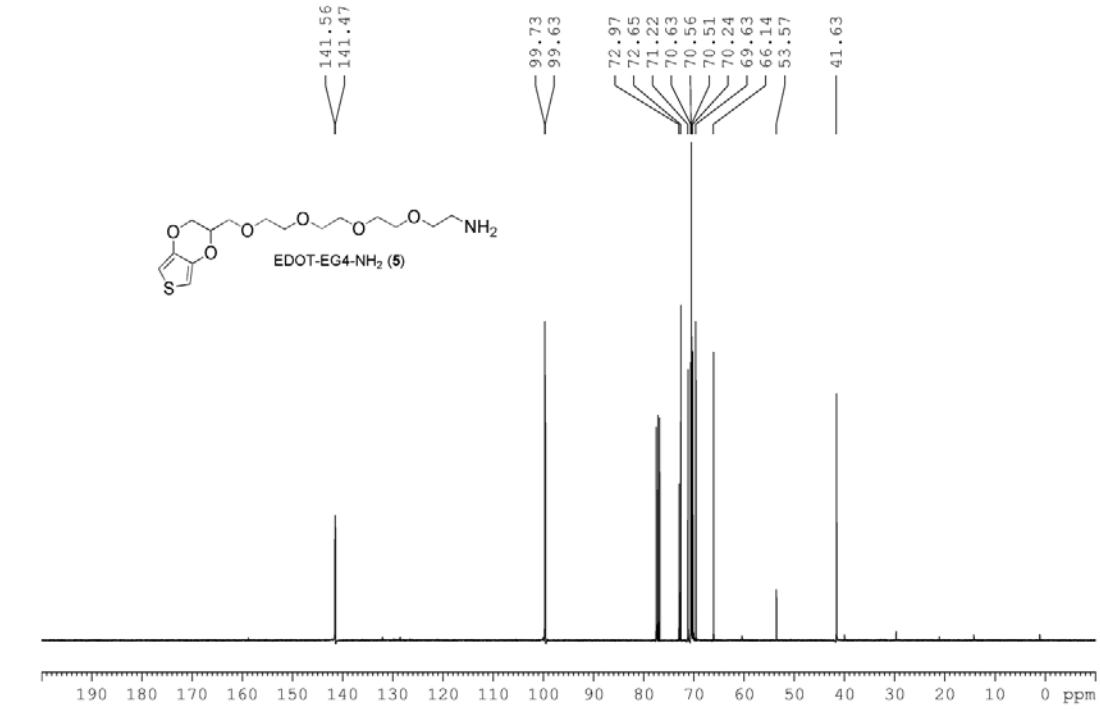


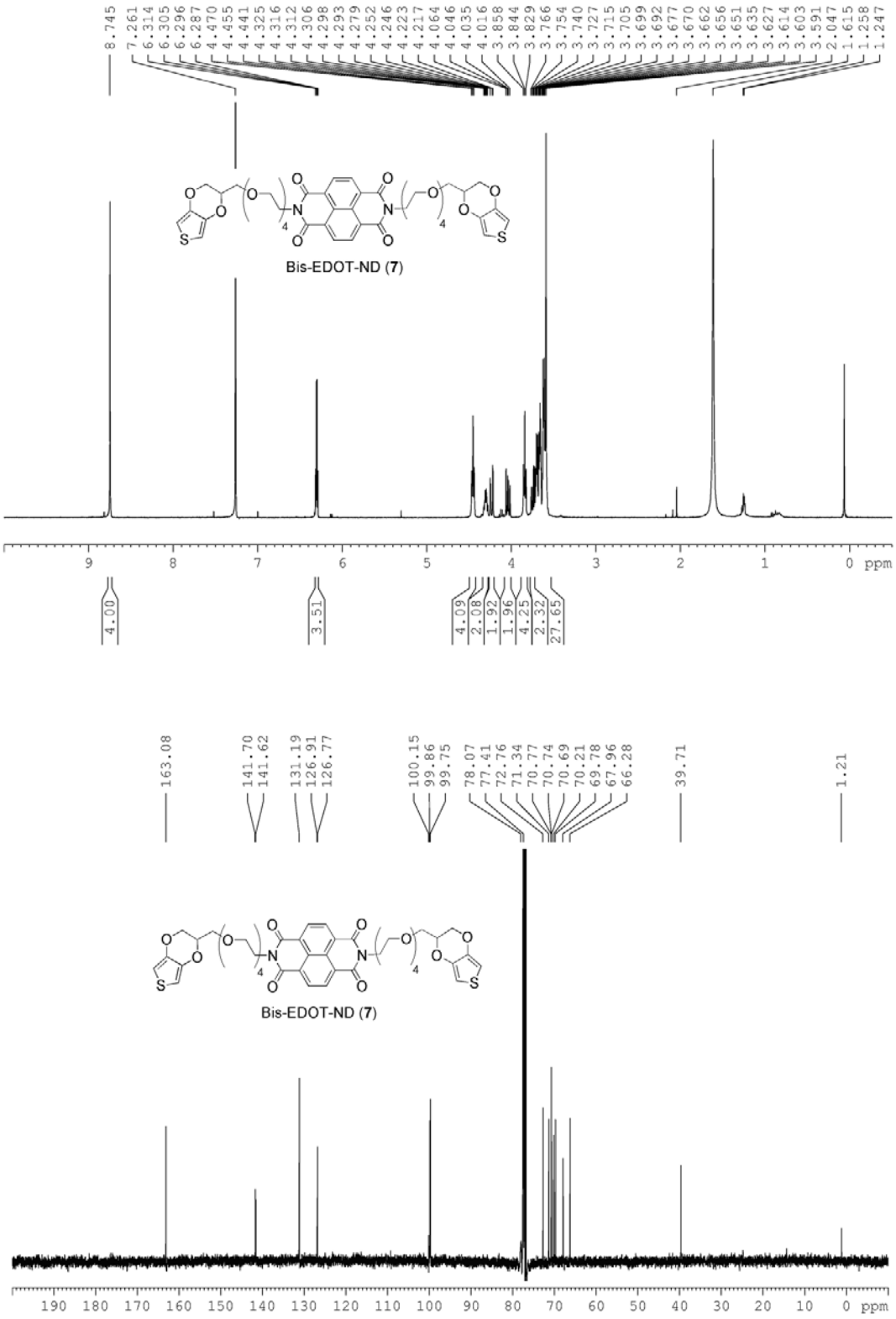

7.260
6.336
6.327
6.325
6.316
4.353
4.347
4.338
4.334
4.328
4.326
4.320
4.315
4.307
4.301
4.273
4.267
4.244
4.238
4.086
4.068
4.057
4.039
3.788
3.776
3.766
3.762
3.749
3.739
3.731
3.711
3.707
3.697
3.692
3.686
3.671
3.660
3.659
3.650
3.647
3.276
3.258
3.241
EDOT-EG4-I (3)
9
8
7
6
5
4
3
2
1
0 ppm
2.00
1.13
1.09
1.10
3.52
14.70
2.25


141.67
141.59
99.89
99.77
72.75
72.10
71.33
70.79
70.73
70.67
70.36
69.76
66.27
53.67
EDOT-EG4-I (3)
190
180
170
160
150
140
130
120
110
100
90
80
70
60
50
40
30
20
10
0 ppm


7.260
6.333
6.323
6.321
6.312
5.299
4.349
4.344
4.335
4.330
4.325
4.322
4.316
4.312
4.303
4.298
4.270
4.264
4.241
4.235
4.083
4.065
4.054
4.035
3.785
3.772
3.758
3.746
3.707
3.702
3.694
3.688
3.680
3.667
3.661
3.654
3.645
3.642
3.399
3.386
3.374
2.959
2.884
1.709
EDOT-EG4-N3 (4)
9 8 7 6 5 4 3 2 1 0 ppm
2.00
1.09
1.09
1.09
1.13
16.85
2.21
0.45
0.44
2.33
141.71
141.63
99.86
99.76
77.41
72.77
71.37
70.86
70.83
70.79
70.69
70.21
69.78
66.30
50.81
EDOT-EG4-N3 (4)
190 180 170 160 150 140 130 120 110 100 90 80 70 60 50 40 30 20 10 0 ppm


7.260
6.295
6.286
6.281
6.272
5.266
4.310
4.305
4.296
4.292
4.286
4.283
4.277
4.272
4.264
4.259
4.229
4.224
4.200
4.194
4.042
4.023
4.013
3.994
3.741
3.729
3.715
3.702
3.667
3.663
3.656
3.653
3.648
3.642
3.626
3.624
3.619
3.615
3.607
3.602
3.591
3.586
3.582
3.578
3.575
3.571
3.566
3.482
3.469
3.456
NH2
EDOT-EG4-NH2 (5)
9 8 7 6 5 4 3 2 1 0 ppm
2.01
1.17
0.47
0.63
1.15
1.28
14.76
2.17
2.09
3.39


141.56
141.47
99.73
99.63
72.97
72.65
71.22
70.63
70.56
70.51
70.24
69.63
66.14
53.57
41.63
NH2
EDOT-EG4-NH2 (5)
190 180 170 160 150 140 130 120 110 100 90 80 70 60 50 40 30 20 10 0 ppm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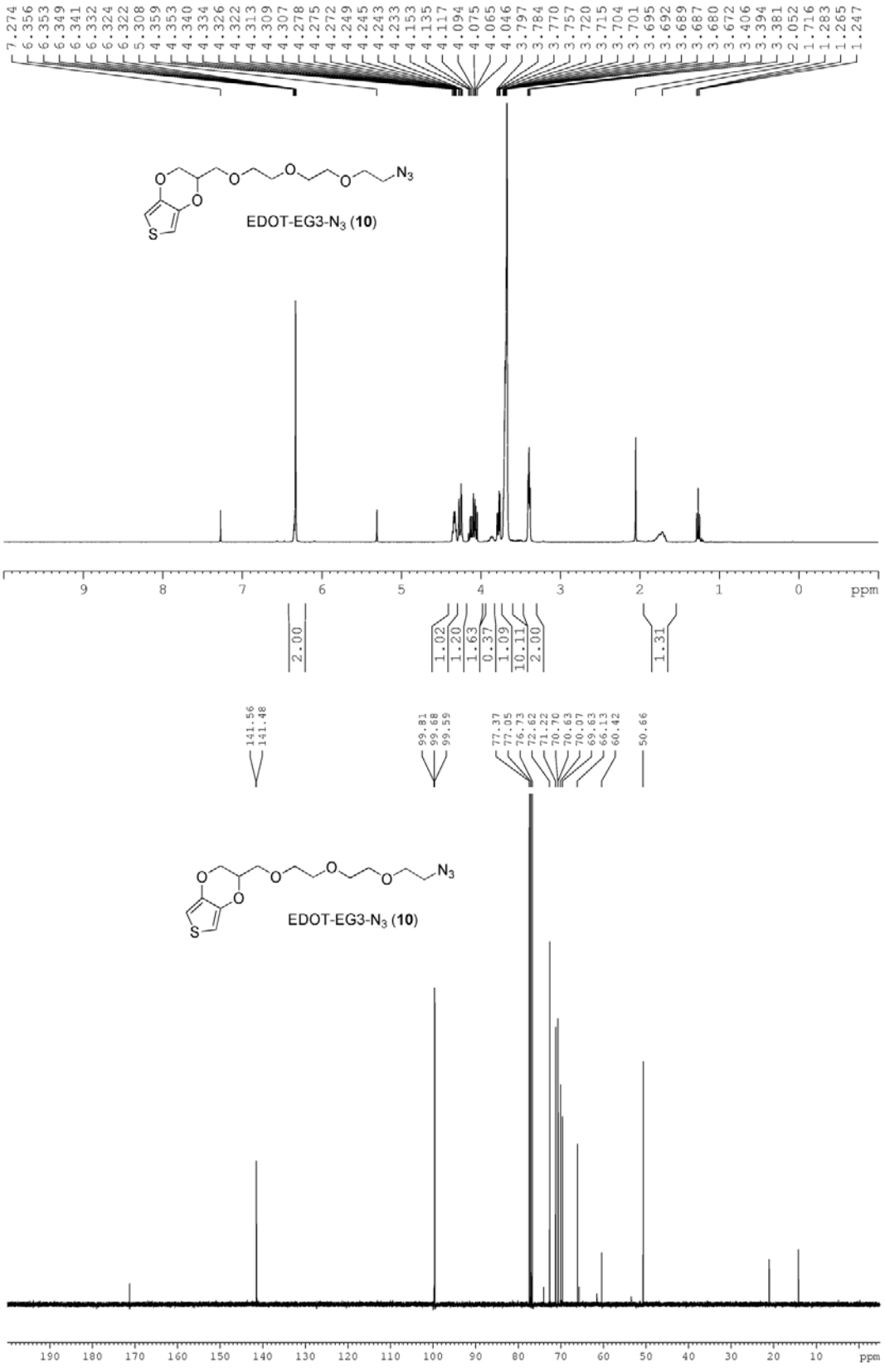
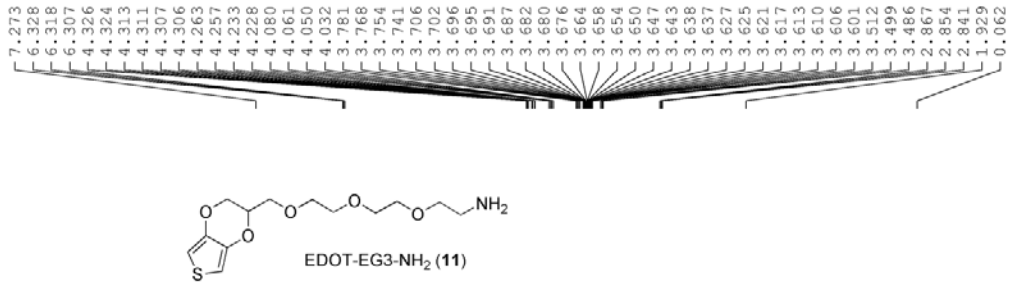
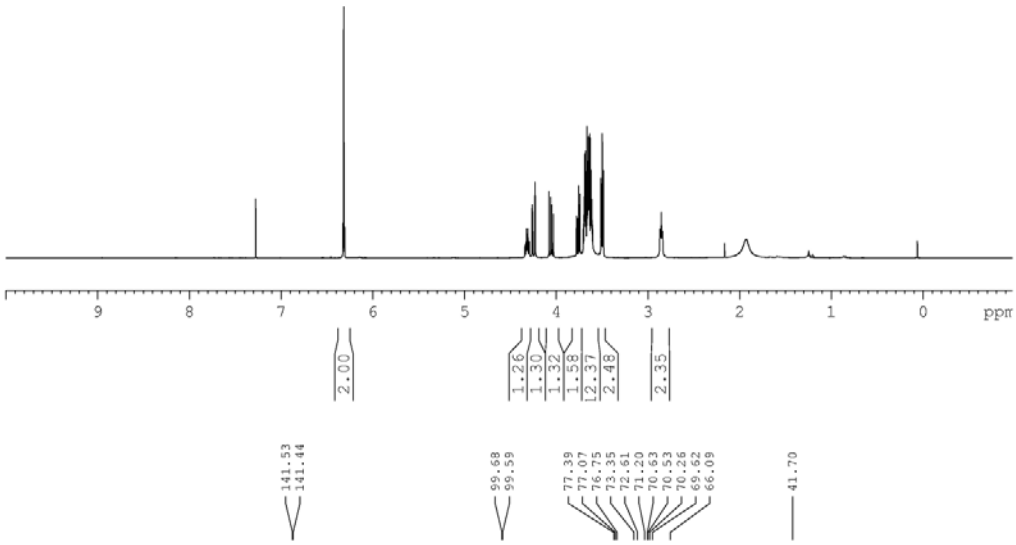
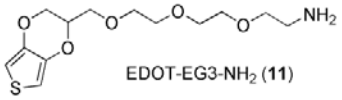
7.261
6.314
6.305
6.296
6.287
4.470
4.455
4.441
4.325
4.316
4.312
4.306
4.298
4.293
4.279
4.252
4.246
4.223
4.217
4.064
4.046
4.035
4.016
3.858
3.844
3.829
3.766
3.754
3.740
3.727
3.715
3.705
3.699
3.692
3.677
3.670
3.662
3.656
3.651
3.635
3.627
3.614
3.603
3.591
2.047
1.615
1.258
1.247
Bis-EDOT-ND (7)
9 8 7 6 5 4 3 2 1 0 ppm
4.00
3.51
4.09
2.08
1.92
1.96
4.25
2.32
27.65
163.08
141.70
141.62
131.19
126.91
126.77
100.15
99.86
99.75
78.07
77.41
72.76
71.34
70.77
70.74
70.69
70.21
69.78
67.96
66.28
39.71
1.21
Bis-EDOT-ND (7)
190 180 170 160 150 140 130 120 110 100 90 80 70 60 50 40 30 20 10 0 ppm


7.274
6.356
6.353
6.349
6.341
6.332
6.324
6.322
5.308
4.359
4.353
4.340
4.334
4.326
4.322
4.313
4.309
4.307
4.278
4.275
4.272
4.249
4.245
4.243
4.233
4.153
4.135
4.117
4.094
4.075
4.065
4.046
3.797
3.784
3.770
3.757
3.720
3.715
3.704
3.701
3.695
3.692
3.689
3.687
3.680
3.672
3.406
3.394
3.381
2.052
1.716
1.283
1.265
1.247
EDOT-EG3-N3 (10)
ppm
2.00
1.02
1.20
1.63
0.37
1.09
10.11
2.00
1.31
141.56
141.48
99.81
99.68
99.59
77.37
77.05
76.73
72.62
71.22
70.70
70.63
70.07
69.63
66.13
60.42
50.66
EDOT-EG3-N3 (10)
ppm

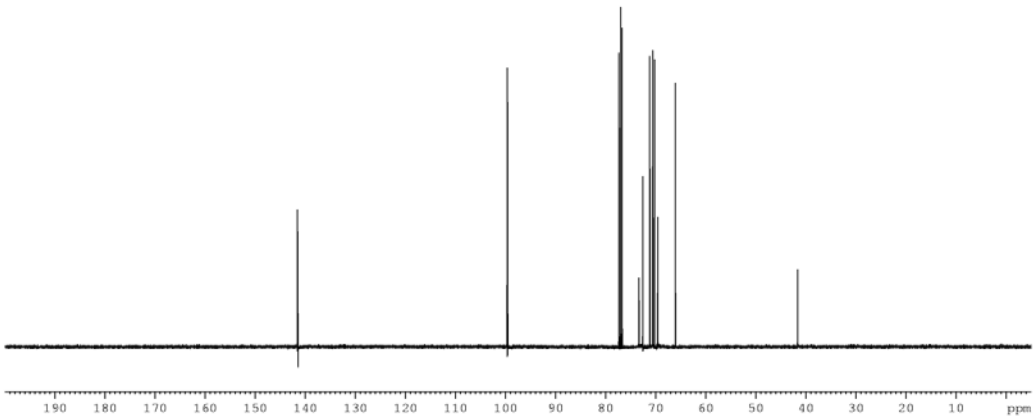


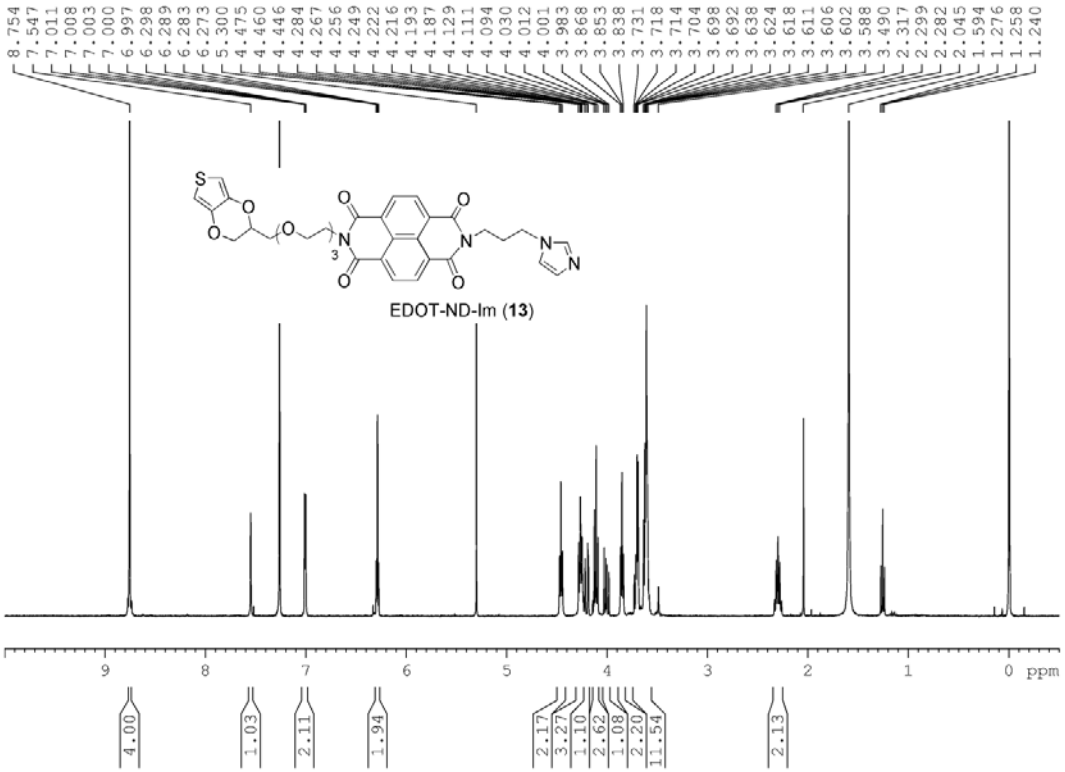
EDOT-EG3-NH$_2$ (**11**)

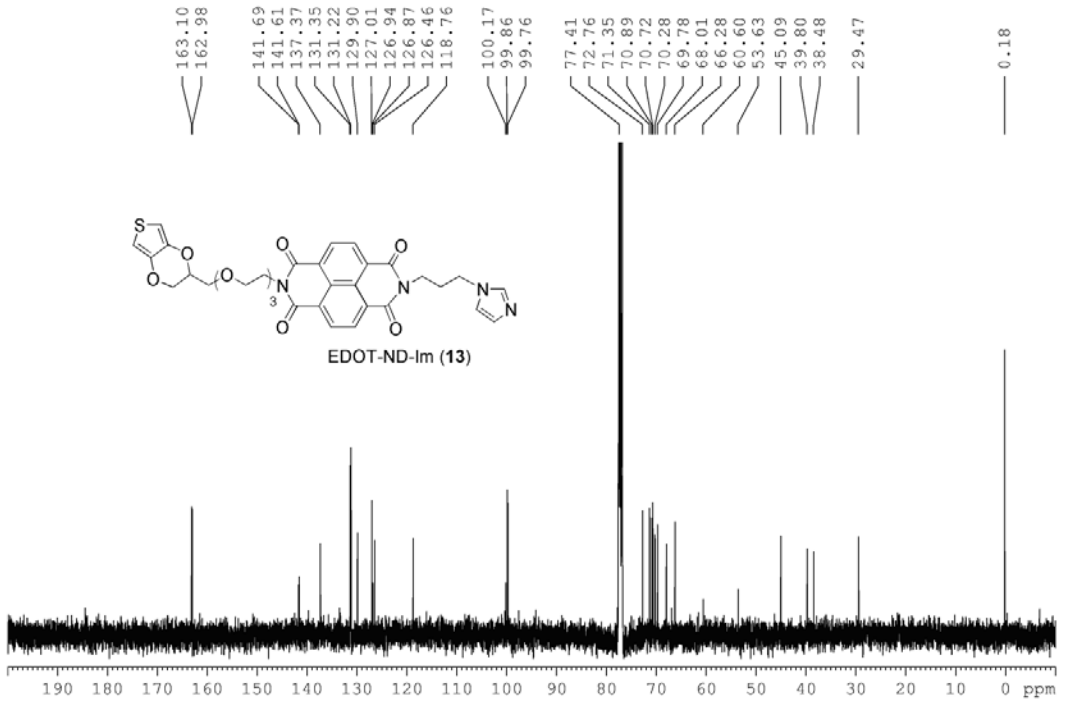




EDOT-EG3-NH$_2$ (**11**)




8.754
7.547
7.011
7.008
7.003
7.000
6.997
6.298
6.289
6.283
6.273
5.300
4.475
4.460
4.446
4.284
4.267
4.256
4.249
4.222
4.216
4.193
4.187
4.129
4.111
4.094
4.030
4.012
4.001
3.983
3.868
3.853
3.838
3.731
3.718
3.714
3.704
3.698
3.692
3.638
3.624
3.618
3.611
3.606
3.602
3.588
3.490
2.317
2.299
2.282
2.045
1.594
1.276
1.258
1.240
EDOT-ND-Im (13)
9 8 7 6 5 4 3 2 1 0 ppm
4.00
1.03
2.11
1.94
2.17
3.27
1.10
2.62
1.08
2.20
11.54
2.13


163.10
162.98
141.69
141.61
137.37
131.35
131.22
129.90
127.01
126.94
126.87
126.46
118.76
100.17
99.86
99.76
77.41
72.76
71.35
70.89
70.72
70.28
69.78
68.01
66.28
60.60
53.63
45.09
39.80
38.48
29.47
0.18
EDOT-ND-Im (13)
190 180 170 160 150 140 130 120 110 100 90 80 70 60 50 40 30 20 10 0 ppm