CHAPTER II

EXPERIMENTS

2.1 Chemicals

Thin layer chromatography (TLC) was performed on aluminium sheets precoated with silica gel (Merck Kieselgel 60 F254) (Merck KgaA, Darmstadt, Germany). Column chromatography was performed using silica gel 0.06-0.2 mm or 70-230 mesh ASTM (Merck Kieselgel 60 G, Merck KgaA, Darmstadt, Germany). Solvents used in synthesis were reagent or analytical grades. Solvents used in column chromatography were distilled from commercial grade prior to use. Other reagents were purchased from the following venders:

- RCI Labscan (Bangkok, Thailand): chloroform, dimethylsulfoxide (DMSO), acetonitrile, acetone AR Grade, dichloromethane AR Grade, dimethylformamide (DMF), sodium hydrogen carbonate ($NaHCO_3$)
- Acrös Organics (USA): 1,2-dibromoethane, 2-bromoethanol, 2,3-dichloroquinoxaline, 1,5-dibromopentane
- Carlo Erba (Milan, Italy): potassium carbonate (K_2CO_3), sodium sulfide (Na_2S), sodium azide (NaN_3), ferric chloride ($FeCl_3$)
- Fluka Chemical (Buchs, Switzerland): sodium metal, diethyl oxalate $(CO_2Et)_2$, triethylamine (TEA), tert-butyl alcohol (t-BuOH), methanesulfonic acid
- Merck Co. (Darmstadt, Germany): ethanol absolute (EtOH), , sodium hydroxide (NaOH), chloroacetyl chloride, concentrated hydrochloric acid (HCl)
- Cambridge Isotope Laboratories, (USA): deuterated chloroform ($CDCl_3$), deuterated dimethylsulfoxide (DMSO-d_6), deuterated acetone (Acetone-d_6)
- Aldrich (USA): propargyl bromide, copper (II) acetate, diethyl oxalate, 4-(dimethylamino)pyridine (DMAP), malonyl chloride, benzyl bromide, phenyl acetylene
- Sigma (USA): (+)-sodium L-ascorbate

- Panreac (Spain): anhydrous magnesium sulfate ($MgSO_4$)
- Ajax Finechem (Auckland, New Zealand): calcium chloride

2.2 Instruments and equipments

Melting points were determined with a Stuart Scientific Melting Point SMP1 (Bibby Sterlin Ltd., Staffordshire, UK). The ^1H NMR and ^{13}C NMR spectra were recorded on a Varian Mercury NMR spectrometer operated at 400.00 MHz for ^1H and 100.00 MHz for ^{13}C nuclei (Varian Company, USA). Deuterated chloroform ($CDCl_3$), deuterated dimethylsulfoxide (DMSO-d_6) or deuterated acetone (Acetone-d_6) were used as the solvent. IR spectra were recorded on a Nicolet 6700 FT-IR RXI spectrometer (Perkin Elmer Instruments, U.S.A.). Mass spectra were recorded on: Waters Micromass Quatto micro API ESCi Mass Spectrometer (Waters, USA). Average molecule weights (M_w and M_n) and polydispersity indices (M_w/M_n) of the polymer were estimated from the measurements on a Waters 2414 gel permeation chromatography (GPC) system, using a set of monodisperse polystyrene as calibration standards. Mass spectra were performed on a Matrix Assisted Laser Desorption/Ionization Time of Fight (MALDI-TOF): Microflex mass spectrometer (Bruker Daltonik GmbH, Germany). The instrument was equipped with a nitrogen laser to desorbed ionize the samples are deposited. α-Cyano-4-hydroxycinnamic acid (CCA) matrix solution for polymer was prepared in 1:2 acetonitrile/H_2O. The samples were dissolved in 0.1%TFA. The UV-Vis absorption spectra were recorded on UV-VISIBLE Spectrometer: UV-2550 (Shimadzu Corporation, Kyoto, Japan). The thermal behaviours of samples were studied by a Mettler Toledo DSC822^e. The specimens of about 7±1 mg were heated under nitrogen atmosphere from 25 to 350 °C at a scan rate of 20 °C.min^{-1}. The temperature was then maintained at 350 °C for 5 min before cooling to 25 °C at the same rate.

2.3 Monomer synthesis

The structures of three dialkyne and three diazide monomers to be synthesized were showed in **Figure 2.1**.

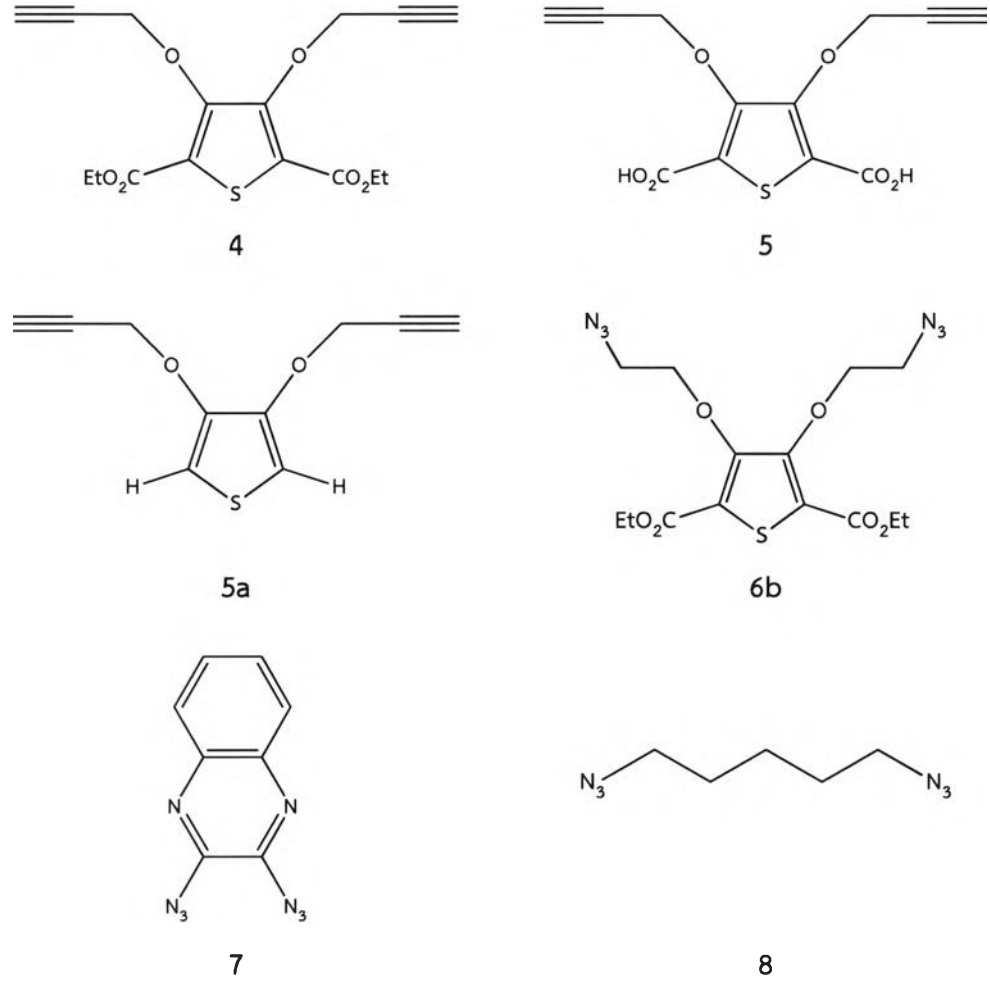
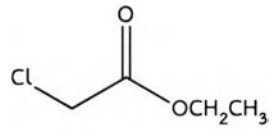


Figure 2.1 The structure of monomers in this research

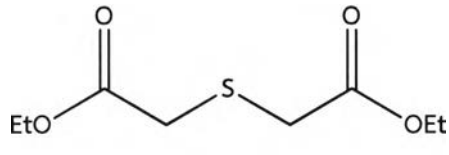
2.3.2 Ethyl chloroacetate (1)



1

An addition funnel containing chloroacetyl chloride (50.000 mL, 6 mmol) was added dropwise into 32 mL ethanol over the period of 30 min at 0 °C. The reaction mixture was stirred at 0 °C for 2.5 h. Then 120 mL of 2 M NaOH was added and the mixture was extracted with ethyl acetate. The organic layer was dried over anhydrous magnesium sulfate and filtered. The solvent was removed on a rotary evaporator to give the product as colorless liquid (50.000 g, 99%). ^{1}H NMR (400 MHz, $CDCl_3$): δ (ppm) 4.24 (q, 2H), 4.05 (s, 2H), 1.30 (t, 3H) **(Figure A.1, Appendix A)**. ^{13}C NMR (101 MHz, $CDCl_3$): δ (ppm) 167.2, 62.1, 40.8, 13.9 **(Figure A.2, Appendix A)**.

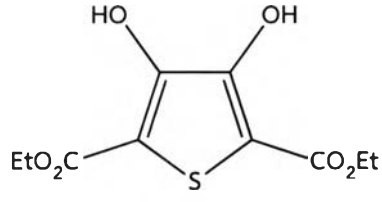
2.3.3 Diethyl thiodiglycolate (2)



2

Sodium sulfide (12.000 g, 50 mmol) was dissolved in 50 mL water and then added dropwise over the period of 30 min to the solution of compound **1** (13.200 g, 55 mmol) in 50 mL acetone. The reaction mixture was stirred at 60 °C under nitrogen for additional 3 h and then extracted with ethyl acetate. The organic layer was dried over anhydrous magnesium sulfate and filtered. The solvent was removed on a rotary evaporator to give the product as yellow liquid. (5.750 g, 60%) ^{1}H NMR (400 MHz, $CDCl_3$): δ (ppm) 4.09 (q, 4H), 3.28 (s, 4H), 1.17 (t, 6H) **(Figure A.3, Appendix A)**. ^{13}C NMR (101 MHz, CDCl3): δ (ppm) 169.5, 61.2, 40.8, 13.9 **(Figure A.4, Appendix A)**.

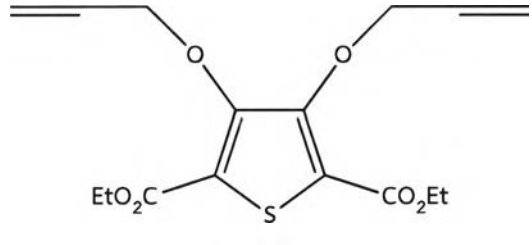
2.3.4 Diethyl 3,4-dihydroxythiophene-2,5-dicarboxylate (3)



3

Sodium metal (2.480 g, 100 mmol) was dissolved in 75 mL ethanol and then added the solution of compound **2** (2.010 g, 11 mmol) and diethyl oxalate (4.540 g, 30 mmol) in 75 ml ethanol dropwise over period of 30 min at 0 °C. The reaction mixture was stirred at 80 °C under nitrogen for 2.5 h, cooled to room temperature and then poured into 100 mL water and acidified with concentrated hydrochloric acid. The resulted precipitate was filtered and vacuum-dried to give the product as a white solid. (1.600 g, 55%) ^{1}H NMR (400 MHz, $CDCl_3$): δ (ppm) 9.37 (s, 2H), 4.40 (q, 4H), 1.39 (t, 6H) **(Figure A.5, Appendix A)**. ^{13}C NMR (100 MHz, $CDCl_3$): δ (ppm) 165.5, 151.6, 107.1, 61.7, 14.0 **(Figure A.6, Appendix A)**. IR (ATR, cm^{-1}): 3293 (O–H st), 2987 (C–H st), 1661 (C=O st), 1508 (C=C st) **(Figure A.7, Appendix A)**. MS: $[M-H]^+$ m/z = 259.20 **(Figure A.8, Appendix A)** [38].

2.3.5 Diethyl 3,4-bis(2-propynyloxy) thiophene-2,5-dicarboxylate (4)

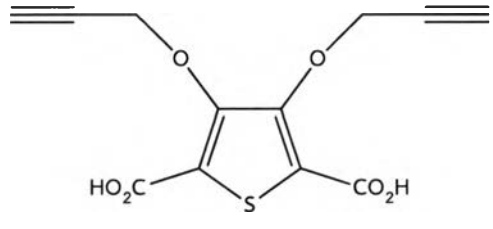


4

Compound **3** (0.800 g, 3 mmol) was dissolved in 5 mL acetonitrile and 2 mL dimethylformamide under nitrogen. The solution was added dropwise to potassium

carbonate (2.070 g, 15 mmol), 4-dimethylaminopyridine (0.030 g, 0.3 mmol), propargyl bromide (2.050 mL of 80% solution in toluene, 18 mmol) and 5 mL acetronitrile over period of 30 min at 80 °C. The reaction mixture was stirred at 80 °C for 4 h. After filtration and evaporation, the crude mixture was purified by silica gel column chromatography, eluting with a 7:3 mixture of hexane and ethyl acetate, giving the product as yellow solid (0.767 g, 76%). mp. 129-131 °C ^{1}H NMR (Acetone-d_6): δ (ppm) 4.88 (d, 4H), 4.23 (q, 4H), 2.99 (s, 2H), 1.23 (t, 6H) **(Figure A.9, Appendix A)**. ^{13}C NMR (Acetone-d_6): δ (ppm) 160.1, 152.3, 121.4, 79.1, 78.1, 62.2, 62.1, 14.4 **(Figure A.10, Appendix A)**. IR (ATR, cm^{-1}): 3304 (≡C–H st), 2980 (C–H st), 2127 (C≡C st), 1707 (–C=O st) **(Figure A.11, Appendix A)**. MS: $[M+Na]^+$ m/z = 359.07 **(Figure A.12, Appendix A)**.

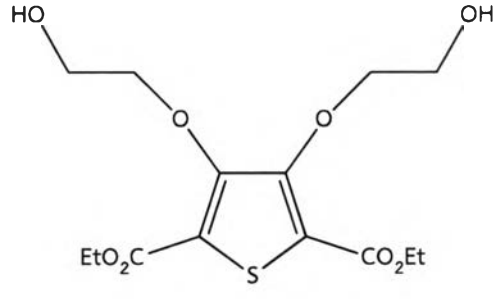
2.3.6 Synthesis of diethyl-3,4-bis(2-propynyloxy)thiophene-2,5-dicarboxylic acid (5)



5

Compound **4** (0.340 g, 1 mmol) and 10 mL 1 M NaOH were mixed with 1 mL ethanol. The reaction mixture was stirred at 80 °C for 4 h, cooled to room temperature and acidified by concentrated hydrochloric acid. The resulted precipitate was then filtered and vacuum-dried to give the product as light gray solid. (0.240 g, 84%) ^{1}H NMR (400 MHz, acetone-d_6): δ (ppm) 4.93 (d, 4H), 2.96 (s, 2H) **(Figure A.13, Appendix A)**. ^{13}C NMR (101 MHz, Acetone-d_6): δ (ppm) 161.5, 152.3, 121.4, 79.2, 78.1, 62.0 **(Figure A.14, Appendix A)**. IR (ATR, cm^{-1}): 3302 (≡C–H st), 2829 (O–H st), 2128 (C≡C st), 1652 (C=O st), 1280 (C–O st) **(Figure A.15, Appendix A)** [39].

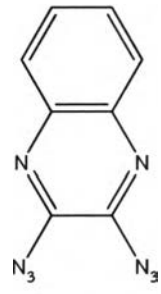
2.3.7 Diethyl 3,4-bis(2-hydroxyethoxy)thiophene-2,5-dicarboxylate (6)



6

Compound **3** (0.540 g. 2 mmol) was dissolved in 2 mL dimethylformamide under nitrogen. The solution was added 2-bromoethanol (1.42 mL, 20 mmol), triethylamine (2.79 mL, 20 mmol) and potassium carbonate (0.55 g, 4 mmol). The reaction mixture was stirred at 130 °C for 25.5 h and then washed with 10% hydrochloric acid and extracted with ethyl acetate. The organic layer was washed with water (5 times). The organic layer was dried over anhydrous magnesium sulfate and evaporated. The crude mixture was purified by silica gel column chromatography, eluting with ethyl acetate, giving the product as white solid. (0.560 g, 74%) ^{1}H NMR (400 MHz, $CDCl_3$): δ (ppm) 4.44 (t, 4H), 4.36 (q, 4H), 3.83 (t, 4H), 1.39 (t, 6H) **(Figure A.16, Appendix A)**. ^{13}C NMR (101 MHz, $CDCl_3$): δ (ppm) 160.9, 153.1, 120.0, 76.5, 61.9, 61.0, 14.0 **(Figure A.17, Appendix A)**. IR (ATR, cm^{-1}): 3335 (O–H st), 2937 (C–H st), 1712 (C=O st), 1486 (C–O st) **(Figure A.18, Appendix A)**. MS: $[M+Na]^+$ m/z = 371.16 **(Figure A.19, Appendix A)**.

2.3.8 Synthesis of 2,3-diazidoquinoxaline (7)



7

2,3-Dichloroquinoxaline (0.390 g, 2 mmol) and sodium azide (0.260 g, 4 mmol) were mixed with 10 mL dimethylformamide. The reaction mixture was stirred at 150 °C for 1 h. After cooling back to room temperature, it was added ethyl acetate and washed with water (5 times). The organic layer was dried over anhydrous magnesium sulfate and filtered. The solvent was removed on a rotary evaporator to give the product as yellow solid. (0.312 g, 72%) ^{1}H NMR (400 MHz, Acetone-d_6): δ (ppm) 8.72 – 8.64 (m, 2H), 8.04 – 7.96 (m, 2H) **(Figure A.21, Appendix A)**. ^{13}C NMR (101 MHz, Acetone-d_6): δ (ppm) 141.8, 131.8, 123.7, 118.8 **(Figure A.22, Appendix A)**. IR (ATR, cm^{-1}): 3075, 3055 (C–H st) 2153 (–N_3 st) **(Figure A.23, Appendix A)**. MS: $[M+Na]^+$ m/z = 235 **(Figure A.24, Appendix A)**.

2.3.9 Synthesis of 1,5-diazidopentane (8)

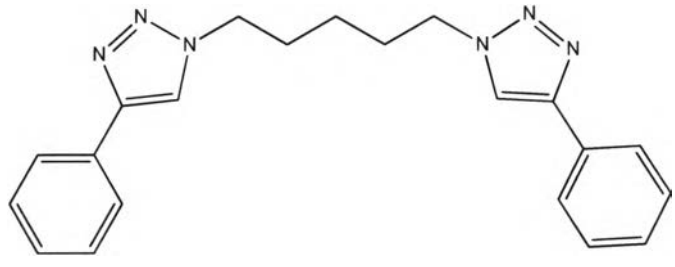
8

1,5-dibromopentane (1.00 mL, 7 mmol) and sodium azide (1.240 g, 19 mmol) were mixed with 10 mL dimethylformamide. The reaction mixture was stirred at room temperature for 24 h and then added ethyl acetate and washed with water (5 times). The organic layer was dried over anhydrous magnesium sulfate and filtered. The solvent was removed on a rotary evaporator to give the product as yellow oil (0.900 g, 83%). ^{1}H NMR ($CDCl_3$): δ (ppm) 3.29 (t, 4H), 1.70–1.56 (m, 4H), 1.53–1.40 (m, 2H)

(Figure A.25, Appendix A). ^{13}C NMR (101 MHz, $CDCl_3$): δ (ppm) 51.2, 28.2, 23.9 **(Figure A.26, Appendix A)**. IR (ATR, cm^{-1}): 2935 (C–H st), 2084 ($-N_3$ st) **(Figure A.27, Appendix A)** [40].

2.4 CuAAC click reaction of diazide

2.4.1 Click reaction synthesis of 1,5-bis(4-phenyl-1,2,3-triazolyl)pentane (9)

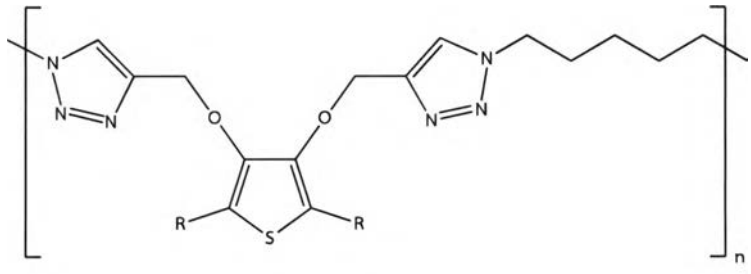


9

Compound **8** (0.500 g, 3.25 mmol) and copper (II) acetate (0.320 g, 1.62 mmol) were mixed in 7 mL tert-butyl alcohol (t-BuOH). Phenylacetylene (8.91 mL, 8.11 mmol) was added into the solution mixture under nitrogen. The reaction mixture was stirred at room temperature for 2 d. After adding 20 mL water, the solution mixture was extracted with ethyl acetate. The organic layer was dried over anhydrous magnesium sulfate and filtered. The solvent was removed on a rotary evaporator to give the product as white solid (0.430 g, 37 %). ^{1}H NMR (400 MHz, $CDCl_3$): δ (ppm) 7.82 (d, 8H), 7.73 (s, 2H), 7.42 (t, 8H), 7.34 (d, 4H), 4.41 (t, 4H), 2.10 – 1.96 (m, 4H), 1.42 (s, 2H) **(Figure A.28, Appendix A)**. ^{13}C NMR (101 MHz, $CDCl_3$): δ (ppm) 132.5, 130.5, 128.8, 128.2, 125.7, 119.6, 49.9, 29.5, 23.4 **(Figure A.29, Appendix A)**. IR (ATR, cm^{-1}) 3119, 3086 (C–H st), 2932 (C–H st), 1463 (C=C st) **(Figure A.30, Appendix A)**.

2.5 Polymer synthesis

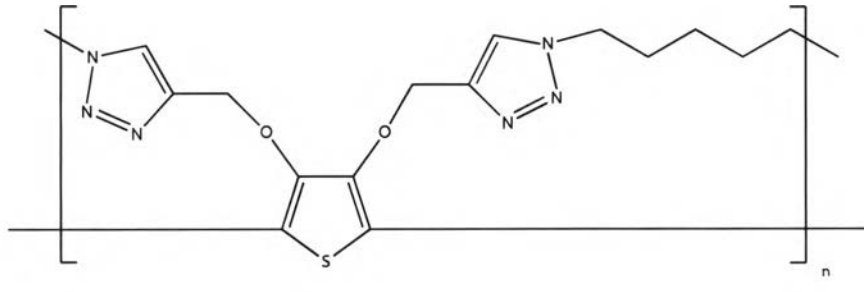
The structures of all polymers (**P1**-**P4**) were showed in **Figure 2.2**.



P1 (R = $CO_2CH_2CH_3$)

P2 (R = CO_2H)

P3 (R = H)



P4

Figure 2.2 The structures of all targeted polymers

2.5.1 Click polymerization to polymer P1

Compound **4** (0.420 g, 125 mmol) was dissolved in 5 mL acetonitrile and then added compound **8** (0.150 g, 100 mmol) and copper (II) acetate (0.200 g, 100 mmol). The reaction mixture was stirred at 0 °C for 6 h and at room temperature for another 42 h. It was added 15 mL dichloromethane and washed with 5 mL water. The organic layer was dried over anhydrous magnesium sulfate and filtered. The solvent was removed on a rotary evaporator to give the product as brown liquid (0.130 g, 27%). ^{1}H NMR ($CDCl_3$): δ (ppm) 7.93–7.78 (2H), 5.71–5.03 (4H), 4.29 (8H), 1.94 (4H), 1.34 (8H)

(Figure A.31, Appendix A). IR (ATR, cm^{-1}): 1703 (C=O st), 1264 (C–O st) **(Figure A.32, Appendix A)**. GPC (polystyrene calibration): M_n = 2903, M_w = 4484; M_w/M_n = 1.54 **(Figure A.33, Appendix A)**.

The synthesis was repeated following the above procedure but kept the reaction stirring at 0 °C for 7 d. The product was obtained as brown liquid (0.200 g, 32%) with the same characterization data as obtained previously. GPC (polystyrene calibration): M_n = 2045, M_w = 3095; M_w/M_n = 1.51 **(Figure A.34, Appendix A)**.

2.5.2 Click polymerization to Polymer P2

Compound **8** (0.208 g, 1.25 mmol) and copper (II) acetate (0.250 g, 1.25 mmol) were mixed with 2 mL tert-butyl alcohol at 80 °C. The solution of compound **5** (0.440 g, 1.56 mmol) in 10 mL tert-butyl alcohol was added dropwise over period of 2 h. The reaction mixture was stirred 80 °C for 63 h and then added dichloromethane and washed with water. The precipitate was then filtered and vacuum-dried to give the product as brown solid (0.281 g, 48 %) ^{1}H NMR (400 MHz, DMSO-d_6): δ (ppm) 8.12 (s, 2H), 5.22 (d, 4H), 4.27 (s, 4H), 1.75 (s, 4H), 1.09 (s, 2H) **(Figure A.35, Appendix A)**. IR (ATR, cm^{-1}): 3142 (O–H st), 2930 (C–H st), 1704 (C=O st) **(Figure A.36, Appendix A)**.

2.5.3 Hydrolysis of polymer P1 to polymer P2

Polymer **P1** (0.509 g, 1.04 mmol) and 10 mL 1 M NaOH were mixed with 2 mL ethanol. The reaction mixture was stirred at 80 °C for 17 h, cooled to room temperature and acidified by concentrated hydrochloric acid. The precipitate was then filtered and vacuum-dried to give the product as light gray solid. (0.312 g, 69%) ^{1}H NMR (400 MHz, DMSO-d_6): δ (ppm) 8.04 (s, 2H), 5.22 (d, 4H), 4.25 (s, 4H), 1.73 (s, 4H), 1.06 (s, 2H) **(Figure A.37, Appendix A)**. IR (ATR, cm^{-1}): 2884 (COO–H st), 1774 (C=O st) **(Figure A.38, Appendix A)**.

2.5.4 Decarboxylation of polymer P2 to polymer P3

Polymer **P2** (0.312 g, 0.72 mmol), cuprous oxide (0.030 g, 0.16 mmol) and quinoline (0.42 mL, 3.59 mmol) were mixed with 10 mL DMSO. The reaction mixture was stirred at 190 °C for 3.5 d, cooled to room temperature and then added water. The precipitate was then filtered and vacuum-dried to give the product as brown solid (0.112 g, 45%) ^{1}H NMR (400 MHz, DMSO-d_6): δ 8.25 (s, 2H), 6.63 (s, 2H), 4.98 (s, 4H), 4.38 (d, 4H), 1.75 (s, 4H), 1.11 (s, 2H). **(Figure A.39, Appendix A).** IR (ATR, cm^{-1}): 2931 (C–H st) **(Figure A.40, Appendix A).** Solid-UV: λ_{max} = 700 nm **(Figure A.41, Appendix A).**

2.5.5 Oxidative coupling polymerization of polymer P3 to double strand polymer P4

The solution of polymer P3 (0.073 g, 0.21 mmol) in 7 mL methanesulfonic acid was stirred at room temperature and then added $FeCl_3$ (0.103 g, 0.63 mmol) into the solution. The reaction mixture was allowed to stir for 2 d. After cooling, it was added dropwise 1M $NaHCO_3$ until $FeCl_3$ was dissolved. The resulted precipitate was recovered by vacuum filtration and vacuum-dried to give the product as black solid. (0.052 g, 72 %) ^{1}H NMR (400 MHz, DMSO-d_6): δ (ppm) 8.10 (s, 2H), 5.21 (s, 4H), 4.28 (s, 4H), 1.81 (d, 4H), 1.16 (d, 2H). **(Figure A.42, Appendix A).** IR (ATR, cm^{-1}): 3017, 2931 (C–H st) **(Figure A.43, Appendix A).** λ_{max} = 630 nm **(Figure A.54, Appendix A).**