Supplementary Information

Polyester-*graft*-phosphorylcholine prepared by ring-opening polymerization and click chemistry

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Materials. L-lactide (LA, 98%), ε-caprolactone (CL, 99%), tin(II) 2-ethylhexanoate ($Sn(Oct)_2$, 95 %), 1,6-hexanediol (97%), benzyl alcohol, copper(I) bromide (CuBr, 98%) and N,N,N',N'',N''-pentamethyldiethylenetriamine (PMDETA, 99 %) were purchased from Aldrich. δ-Valerolactone and *n*-butyllithium in hexanes (2.9M) were purchased from Alfa Aesar. Copper(II) sulfate pentahydrate (99.6%, Fisher Scientific) and sodium ascorbate (Source Naturals) were used as received. L-lactide was recrystallized from dry ethyl acetate, then dried under vacuum at room temperature. ε-Caprolactone, 1,6-hexanediol, δ-valerolactone, benzyl alcohol, and dichloromethane were dried and distilled over calcium hydride. L-lactide and 1,6-hexanediol were lyophilized before use. α-Propargyl δ-valerolactone[1] and phosphorylcholine azide[2] were synthesized as previously reported. Copper(I) bromide was purified by stirring in acetic acid, washing with methanol, and drying under vacuum at room temperature. Tetrahydrofuran (THF) was dried and distilled over sodium/benzophenone ketyl.

Instrumentation. Nuclear magnetic resonance (NMR) spectroscopy was performed on Brüker Avance 400 and Spectrospin 300 machines. Molecular weights and polydispersity indices (PDIs) were estimated by gel permeation chromatography (GPC) in THF against polystyrene standards, operating at 1.0 mL/min with a Knauer HPLC pump K-501, refractive index detector K-2301, and three PL gel MixedD columns (5μm, 300 mm x 7.5 mm). Aqueous GPC was performed in 0.1 M sodium nitrate and 0.02 weight percent sodium azide buffer against poly(ethylene oxide) calibration standards, operating at 1.0 mL/ min with an HP Series 1050 Pump, HP 1047A refractive index detector, and three Waters Ultrahydrogel columns (7.8 x 300 mm). Microwave heating was performed using a SmithCreater single-mode microwave, giving continuous radiation of 2.45 GHz (Personal Chemistry, Inc.).

Synthesis of poly(α-propargyl δ-valerolactone) ***2***. 1.7 M stock solutions of benzyl alcohol and tin(II) 2-ethylhexanoate were prepared in dry THF. Aliquots from stock solutions of tin (II) ethyl hexanoate (11 μL, 0.018 mmol) and benzyl alcohol (106 μL, 0.18 mmol) and α-propargyl δ-valerolactone (1.5 g, 11 mmol) were added to a flame-dried Schlenk tube under a stream of nitrogen. The tube was placed under vacuum for 20 minutes at room temperature then was closed under vacuum. The closed Schlenk tube was placed in an oil bath at 100 °C for 28 h. Following cooling to room temperature, the mixture was dissolved in dichloromethane and precipitated repeatedly in cold hexanes. The polymer was isolated by filtration, then dried under vacuum at room temperature overnight. The molecular weight of the polymer was determined by ^{1}H NMR spectroscopy (end-group analysis) in $CDCl_3$ to be 8,700 g/mol. ^{1}H NMR (300 MHz, $CDCl_3$) δ ($CHCl_3$ = 7.26 ppm) 4.04 (m, 2H, C***H*****$_2$**OCO), 2.5 (m, 1H), 2.48 (m, 2H), 2.02 (t, 1H, C≡C***H***), 1.64 (br, 4H) ^{13}C NMR ($CDCl_3$, 100 MHz) 174.1 (CH_2O***C***O), 81.1(***C***≡CH***)***, 70.3(C≡***C***H***)***, 64.5, 44.2, 27.6, 26.3, 21.3 ppm. The GPC estimated molecular weight in THF was 8,300 g/mol and PDI 1.16.

Click reaction of homopolymer ***2*** *with phosphorylcholine azide under microwave conditions*

Phosphorylcholine azide (748 mg, 2.4 mmol) was dissolved in degassed deionized water (3 mL) in a pyrex glass vial equipped with a small magnetic stir bar. To this was added 2 mL of a degassed THF solution of homopolymer (M_n= 8300g/mol, 276 mg, 2.0 mmol of alkyne groups), followed by $CuSO_4$ (24.9 mg, 0.1 mmol) and sodium ascorbate (198 mg, 1 mmol). The tube was sealed, and the mixture was irradiated with 2.45 GHz, maintaining a constant temperature of 70 °C for 5 minutes. The mixture was cooled to room temperature, and the solvent was removed by rotary evaporation, and the product was dissolved in distilled water (10 mL). Stirring with Cuprisorb™ removed the residual copper. The solution was filtered to remove the Cuprisorb™ and dialyzed (MWCO = 1000) for one day. The solution was lyophilized to obtain the desired product (552 mg, 61%). Aqueous GPC: 10,000 g/mol PDI 1.32. ^{1}H NMR (400 MHz, D_2O) δ 7.76 (br, 1H, C=C***H***), 4.29 (br,2H, C=CHNC***H$_2$),*** 4.26 (br, 2H), 4.02 (br, 2H,C***H$_2$***O), 3.79-3.65 (m, 2H), 3.21 (m, 9H N***Me$_3$***) 2.82 (m, 2H, C***H***$_2$C=CH), 2.43(m, 2H), 2.31 (br, 1H), 1.73 (br, 8H), 1.46 (br, 2H), 1.23 (br, 2H), 1.12 (br, 2H); ^{13}C NMR (D_2O, 100 MHz) 178.1 (CH_2O***C***O), 144.5(C=***C***H), 122.1 (***C***=CH), 66.0, 61.2, 59.2, 53.9 (N***Me$_3$***), 49.8, 29.6, 27.6, 25.4, 24.8, 16.5 ppm.

Synthesis of poly(L-lactide-co-ε-caprolactone-co-alkyne valerolactone) ***5***. L-lactide (3.58 g, 24.8 mmol), ε-caprolactone (1.0 mL, 9.0 mmol), and α-propargyl δ-valerolactone (1.6 g, 11.4 mmol) were introduced into a flame-dried Schlenk tube under a stream of nitrogen. A stock solution of 0.6 mL of tin(II) 2-ethylhexanoate in 9.6 mL of dry THF was prepared. 26 μL of this solution (5 μmol of tin(II) 2-ethylhexanoate) was added, followed by 1,6-hexanediol (6 mg, 52 μmol). The Schlenk tube was placed in an oil bath at 70 °C under vacuum for 15 min, and purged by a short release under nitrogen. This action was repeated three times, after which the Schlenk tube was closed under vacuum. The oil bath temperature was then increased to 170 °C, and the mixture was stirred for 15 h. Following cooling to room temperature, the mixture was dissolved in chloroform and precipitated repeatedly into methanol. The polymer was isolated by filtration, then dried under vacuum at room temperature overnight. The composition of the final polymer (relative ratio of incorporated monomers) was determined by ^{1}H NMR spectroscopy in $CDCl_3$ to be LA:CL:AVL = 60:20:20. ^{1}H NMR (400 MHz $CDCl_3$) δ (C*H*Cl_3 = 7.26 ppm) 5.12 (m, 3H, C***H***$_3$ LA), 4.07 (m, 4H, C***H*****$_2$**OCO), 2.58 (m, 1H), 2.45 (m,2H), 2.28 (m, 2H), 2.00 (t, 1H, C≡C***H***), 1.45-1.74 (m, 6H) ^{13}C NMR ($CDCl_3$, 100 MHz) 173.8 (CH_2O***C***O, AVL), 172.7 (CH_2O***C***O, CL), 169.5 (CH_2O***C***O, LA), 80.7 (***C***≡CH***)***, 70.2 (C≡***C***H***)***, 68.5, 65.2, 64.1, 43.5, 33.5, 28.2, 28.1, 26.9, 25.1 24.2, 21.1, 16.6 ppm. The GPC estimated molecular weight of the terpolymer in THF was 48,000 g/mol, and PDI 2.19.

Click reaction of terpolymer ***5*** *with phosphorylcholine azide*

The terpolymer (492 mg, 0.70 mmol of alkyne groups) and PC-azide **3** (234 mg, 0.76 mmol) were placed in a two-neck round-bottom flask equipped with a septum and a vigreux column. CuBr (108 mg, 0.75 mmol) and dichloromethane (10 mL) were added, and the mixture was stirred at room temperature until a homogeneous solution was seen. PMDETA (0.32 mL, 1.53 mmol) was added, and the mixture was heated with stirring at 30 °C under nitrogen atmosphere for 24 hours. The reaction mixture was concentrated by rotary evaporation, dissolved in methanol and precipitated in THF. The isolated polymer was dissolved in water and stirred with Cuprisorb™ to remove residual copper. The solution was filtered to remove residual solids, and dialyzed (MWCO = 6000-8000) for one day. The solution was lyophilized to obtain the product (484 mg, 67%). ^{1}H NMR (400 MHz, D_2O) δ 5.11 (m,2H,C***H***CO, LA), 4.32 (m,2H, C=CHNC***H*****$_2$),** 4.27 (m, 2H), 4.04 (br, 4H), 2.84 (m, 2H), 3.67 (m, 2H), 3.23 (s, 9H, NMe_3), 2.87 (m, 2H, C***H***$_2$C=CH), 2.40 (br, 2H), 2.29 (br, 3H),1.81 (br, 8H), 1.25-1.70 (br, 6H), 1.55 (m, 2H, CHC***H***$_3$) ppm. ^{13}C NMR (D_2O, 100 MHz) δ 169.4 (CO), 144.3 (C=***C***H), 122.6 (***C***=CH), 69.0, 65.9, 59.2, 59.1, 53.8 (N***Me*****$_3$**), 50.0, 29.7, 27.5, 25.4, 24.5, 24.4, 16.3, 16.2 ppm.

Cell culture and cytotoxicity assays. Human breast adenocarcinoma cells (MCF7; American Type Culture Collection) were maintained in Eagle's Minimum Essential Medium containing 10% fetal bovine serum and 0.01 mg/mL bovine insulin (Sigma) at 37 °C with 5% CO_2. The MCF7 cells were seeded into sterile 96 well plates at 1 x 10^5 cell density and 48 hours later treated with different concentrations of PC-grafted-polyester suspended in sterile water. Cell viability at 24 and 48 hours incubation was determined using CellTiter-Glo Luminescent Cell Viability Assay (Promega). Following the indicated periods of incubation with different concentrations of PC-grafted polyester, cells were incubated with CellTiter-Glo reagent for 15 minutes, and the luminescence values determined on the FLUOstar OPTIMA Plate reader (BMG LABTECH) and the percentage cell viability were calculated with respect to the untreated control cells. Each plate had at least 8 replicates. Standard error mean (S.E.M) was determined using GraphPad Prism 4 for windows.

1. B. Parrish, R. B. Breitenkamp and T. Emrick, *J. Am. Chem. Soc.*, 2005, **127**, 7404-7410.
2. D. Samanta, K. Kratz, X. Zhang and T. Emrick, *Macromolecules*, 2008, **41**, 530-532.