Supporting Information

Ring-Opening Polymerization of γ-(4-Vinylbenzyl)-$_L$-Glutamate *N*-Carboxyanhydride for the Synthesis of Functional Polypeptides

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Materials All chemicals were purchased from Sigma-Aldrich (St. Louis, MO, USA) and used as received unless otherwise specified. Anhydrous dimethylformamide (DMF) was dried by columns packed with 4Å molecular sieves and stored in a glove-box. Tetrahydrofuran (THF) and hexane were dried by columns packed with alumina and stored in a glove-box. Dry nitrobenzene (NB) was prepared by treating regular nitrobenzene with CaH_2 followed by distillation under reduced pressure. H-Lys(Z)-OH and H-Glu(OBn)-OH were purchased from Chem-Impex International (Des Plaines, IL, USA) and used as received. Glu-NCA and Lys-NCA were prepared by following previously reported procedures.[1]

Instrumentation NMR spectra were recorded on a Varian UI400 MHz, a UI500NB MHz or a VXR-500 MHz spectrometer. Tandem gel permeation chromatography (GPC) experiments were performed on a system equipped with an isocratic pump (Model 1100, Agilent Technology, Santa Clara, CA, USA), a DAWN HELEOS 18-angle laser light scattering detector (also known as

multi-angle laser light scattering (MALLS) detector, Wyatt Technology, Santa Barbara, CA, USA) and an Optilab rEX refractive index detector (Wyatt Technology, Santa Barbara, CA, USA). The detection wavelength of HELEOS was set at 658 nm. Separations were performed using serially connected size exclusion columns (100 Å, 500 Å, 10^3 Å and 10^4 Å Phenogel columns, 5 μm, 300 × 7.8 mm, Phenomenex, Torrance, CA, USA) at 60 °C using DMF containing 0.1 M LiBr as the mobile phase. The MALLS detector was calibrated using pure toluene with no need for external polymer standards and can be used for the determination of the absolute molecular weights. The molecular weights (MWs) of all polymers were determined based on the *d*n/*d*c value of each sample calculated offline by using the internal calibration system processed by the ASTRA V software (version 5.1.7.3, Wyatt Technology, Santa Barbara, CA, USA). Infrared spectra were recorded on a Perkin Elmer 100 serial FTIR spectrophotometer calibrated with polystyrene film. Circular dichroism (CD) measurements were carried out on a JASCO J-720 CD spectrometer (Jasco Analytical Instruments, Easton, MD, USA). Ozone was produced by an OZV-8S ozone generator manufactured by Ozone Solutions Inc (Hull, IA, USA). Lyophilization was performed on a FreeZone lyophilizer (Labconco, Kansas City, MO, USA). UV light was generated from an OmiCure S1000 UV lamp (EXFO, Mississauga, Canada). Small angle X-ray scattering (SAXS) data were collected in a helium chamber using a Bruker M18XHF[22] (Bruker Axs Inc. Madison, WI, USA) rotating anode generator operating at 50kV and 50mA supplying a Cu Kα (λ = 1.541838 Å) radiation beam that was collimated using a pinhole collimator.

Synthesis of γ-(4-vinylbenzyl)-$_L$-glutamate NCA (VB-Glu-NCA) γ-(4-Vinylbenzyl)-$_L$-glutamate (VB-Glu) was synthesized by following a reported procedure with slight modification.[2] γ-(4-Vinylbenzyl)-$_L$-glutamate (2.45 g, 10 mmol) was dried under vacuum for 2 h. This solid was suspended in anhydrous THF (30 mL). Phosgene (20% in toluene, 7 mL) was added under nitrogen to the suspension dropwise over the course of 5 min. The suspension was stirred at 50 ^{o}C for 2-3 h. The solvent was removed under vacuum. The residue was dissolved in anhydrous THF (20 mL) in a glove box and centrifuged to remove the undesired byproducts. The supernatant was combined and the solvent was removed under vacuum. The residue was dissolved in THF (10 mL) followed by the addition of anhydrous ether (100 mL). The solution

was cooled at -30 °C in the box. A dark oily residue sticking to the bottom of the flask was removed by carefully pouring the upper level clear solution into another clear 250mL flask. The clear solution containing NCA was combined and concentrated. VB-Glu-NCA in white crystalline form (3.4 mmol, 1.0 g, 34% yield) was obtained through recrystallization three times using THF/Hexane. ^{1}H NMR ($CDCl_3$, 500 MHz): δ 7.40 (d, *J* =8.0 Hz, ArH, 2H), 7.30 (d, *J* =8.0 Hz, ArH, 2H), 6.73-6.67 (m, NH and C_6H_4CH=CH_2, 2H), 5.76 (d, *J* =17.5 Hz, C_6H_4CH=CH_2, 1H), 5.27 (d, *J* =11 Hz, C_6H_4CH=CH_2, 1H), 5.11 (s, ArCH_2, 2H), 4.38 (t, αH, *J* =6.0 Hz, 1H), 2.58 (t, γH, 2H), 2.26 (m, βH, 1H), 2.12 (m, βH, 1H). ^{13}C NMR ($CDCl_3$, 500 MHz): δ 172.4, 169.3, 151.9, 137.8, 136.1, 134.6, 128.6, 126.4, 114.6, 66.8, 56.9, 29.8, 26.8. ESI MS analysis (with NaCl) Calcd: *m*/*z* 289.2 (M); found: *m*/*z* 312.3 (M+Na). Anal. Calcd. For $C_{15}H_{15}NO_5$: 62.29%C, 5.21%H, 4.84%N; found: 62.06%C, 5.12%H, 4.83%N.

General procedure for the VB-Glu-NCA polymerization In a glove-box VB-Glu-NCA (29 mg, 0.1 mmol) was dissolved in a mixture of DMF (450 μL) and nitrobenzene (30 μL). The VB-Glu-NCA solution was added to a DMF solution of HMDS (20 μL, 0.1 mmol/mL). The reaction mixture was stirred for 15 h at room temperature. An aliquot of the polymerization solution was diluted to 10 mg PVBLG/mL using DMF (containing 0.1 M LiBr), and then analyzed by GPC to determine the M_w and M_n. The *N*-terminus of the polymer was capped by treating the polymerization solution *in situ* with tetrabutylammonium fluoride solution (1.0 M × 50 μL), diisopropylethylamine (30 μL) and benzyl chloroformate (0.1 mL) for 2-3 h. The majority of DMF was then removed under reduced pressure. The residue was precipitated with ether (15 mL). The obtained PVBLG was sonicated for 5 min in ether (5-10 mL) and centrifuged to remove the solvent. After the sonication-centrifugation procedure was repeated two more times, PVBLG was collected, dried under vacuum and stored in -20 °C freezer (isolated yield 85 %). ^{1}H NMR (TFA-*d*, 500 MHz): δ 7.53 (d, *J* =7.0 Hz, ArH, 2H), 7.39 (d, *J* =7.0 Hz, ArH, 2H), 6.84 (dd, J_1 =11.0 Hz, J_2 =18.0 Hz C_6H_4CH=CH_2, 1H), 5.91 (d, *J* =18.0 Hz, C_6H_4CH=CH_2, 1H), 5.43 (d, *J* =11.0 Hz, C_6H_4CH=CH_2, 1H), 5.26 (m, ArCH_2, 2H), 4.80 (m, αH, 1H), 2.68 (m, γH, 2H), 2.30 (m, βH, 1H), 2.12 (m, βH, 1H)

Kinetic study of the polymerization of VB-Glu-NCA In a glove-box, VB-Glu-NCA (172 mg, 0.6 mmol) was dissolved in DMF (3.0 mL). A stock solution of HMDS (0.1 M, 0.06 mL) was added to the stirred VB-Glu-NCA/DMF solution in one portion. The real-time concentration of NCA was quantified by measuring the intensity of the anhydride band at 1784 cm^{-1} by FT-IR. The conversion of NCA was determined by comparing the NCA concentration in the polymerization solution versus the initial NCA concentration.

Reactions of PVBLG (Scheme 1)

Protocol of route a: $PVBLG_{70}$ (45 mg, 0.18 mmol) was dissolved in chloroform (15 mL) at -78 ^{o}C. O_2 was bubbled into the solution for 1 min followed by bubbling of O_3 until the solution became blue, indicating the reaction was complete. O_3 was then replaced with O_2, which was bubbled into the solution over the course of 2 min until the solution became colorless. The solution was then degassed and back filled with nitrogen. $NaBH_4$ (95 mg, 2.5 mmol) was then added to the mixture. The solution was stirred at room temperature overnight and the solvent was removed under reduced pressure. Water (10 mL) was added to the residue. The solution was stirred at room temperature for 1 h. The polymer was collected as a solid by filtration and washed by DI water (5 mL × 2). The resulting $(PVBLG\text{-}a)_{70}$ was freeze dried to give 33 mg white solid in 72% yield. ^{1}H NMR ($CDCl_3$/TFA-*d* =5/1, 500 MHz): δ7.28 (Ar<u>H,</u> 4H), 5.17 (-CO_2<u>CH_2</u>$C_6H_4CH_2$-, 2H), 4.78 (-$CO_2CH_2C_6H_4$<u>CH_2</u>-, 2H), 4.62 (αH, 1H), 2.53 (γH, 2H), 2.17 (βH, 1H), and 1.96 (βH, 1H).

Protocol of route b: $PVBLG_{70}$ (49 mg, 0.2 mmol) was dissolved in chloroform (15 mL) at -78 ^{o}C. O_2 was bubbled into the solution for 1 min followed by bubbling of O_3 until the solution became blue, indicating that the reaction was complete. O_3 was then replaced with O_2, which was bubbled into the solution over the course of 2 min until the solution became colorless. The solution was then degassed and back filled with nitrogen. PPh_3 (131 mg, 0.5 mmol) was then added to the mixture. The solution was stirred at room temperature for 2-3 h. The solvent was removed under reduced pressure. The resulting poly(γ-(4-aldehydebenzyl)-$_L$-glutamate) (PABLG or PVBLG-b) was purified by adding methanol followed by sonication (3 × 15 mL) to remove

the unreacted PPh_3 and other impurities and lyophilized (39 mg, 78% yield). ^{1}H NMR (TFA-*d*, 500 MHz): δ 10.31 (O=C<u>H</u>C_6H_4-, 1H), 8.40 (Ar<u>H</u>, 2H), 7.96 (Ar<u>H</u>, 2H), 5.71 (O=CHC_6H_4<u>CH_2</u>, 2H), 5.21 (αH, 1H), 3.12 (βH, 2H), 2.75 (γH, 1H), and 2.56 (γH, 1H). FT-IR: 1734 cm^{-1}(ester carbonyl), 1704 cm^{-1}(aldehyde carbonyl), 1654 cm^{-1}(Aminde I), and 1547 cm^{-1}(amide II).

Protocol of route c[3]*:* OsO_4 (2.5 wt% in *tert*-butanol, 0.1 mL) and oxone (614 mg, 2 mmol) were added to a solution of $PVBLG_{70}$ (20 mg, 0.08 mmol vinyl) in DMF (1 mL). The mixture was stirred at room temperature for 2 days. The excess oxone was quenched by Na_2SO_3 (200 mg, 1.6 mmol in 5 mL water). The solution was tuned to slightly basic by the addition of 2 M $NaHCO_3$, dialyzed against water, and then lyophilized to give $(PVBLG\text{-}c)_{70}$ as white solid (81% yield). ^{1}H NMR (DMSO-d_6/TFA-*d* = 1/1, 500 MHz): δ7.00-6.90 (broad, Ar<u>H,</u> 4H), 4.80-4.52 (broad, -CO_2<u>CH_2</u>C_6H_4-, 2H), 4.03-3.78 (αH, 1H), 2.5-1.80 (broad, γH and βH, 4H).

Protocol of route d: $PVBLG_{70}$ (25 mg, 0.1 mmol vinyl group), OsO_4 (2.5 wt% in *tert*-butanol, 0.1 mL) and *N*-methylmorpholine *N*-oxide (57 mg, 0.49 mmol) were stirred in acetone/H_2O (v/v = 10:1; 1.5 mL total volume) for 20 h. The excess NMO was treated with Na_2SO_3 (126 mg, 1 mmol in 5 mL water). The solution was purified by dialysis against water and dried by lyophilization to give $(PVBLG\text{-}d)_{70}$ as a white solid (22 mg, 79 % yield), which was analyzed by ^{1}H NMR (DMSO-d_6/TFA-*d* =5/1, 500 MHz): δ7.78 (N<u>H</u>, 1H), 7.42-7.03 (Ar<u>H,</u> 4H), 5.20-4.83 (-CO_2<u>CH_2</u>C_6H_4-, 2H), 4.51 (-$CO_2CH_2C_6H_4$<u>CH</u>(OH)CH_2OH, 1H), 4.22-3.83 (broad, αH, 1H), 3.37 (-$CO_2CH_2C_6H_4CH$(OH)<u>CH_2</u>OH, 2H), 2.5-1.8 (broad, γH and βH, 4H).

Protocol of route e: Grubbs catalyst (2nd generation, 2 mg, ca. 0.0025 mmol) and *cis*-1,4-dichloro-2-butene (0.2 mL, 1.9 mmol) were dissolved in dry CH_2Cl_2 (2 mL) in a glove-box. $PVBLG_{70}$ (25 mg, 0.1 mmol vinyl group) was added to the mixture via syringe. The solution was stirred at room temperature for 24 h. The solvent was removed under vacuum. The residue was washed by ether (2 × 10 mL) and dried under vacuum to give $(PVBLG\text{-}f)_{70}$ as a light yellow oil (23 mg, 78 % yield). ^{1}H NMR ($CDCl_3$/TFA-*d* = 5/1, 500 MHz): δ 7.73-7.21 (broad, Ar<u>H,</u> 4H), 6.63(-C_6H_4<u>C</u>H=CHCH_2Cl, 1H), 6.28 (-C_6H_4CH=<u>CH</u>CH_2Cl, 1H), 5.15

(-$CO_2\underline{CH_2}C_6H_4$-, 2H), 4.62 (αH, 1H), 4.21 (-C_6H_4CH=$CH\underline{CH_2}Cl$, 2H), 2.58 (βH, 2H), 2.18 (γH, 1H), and 1.97 (γH, 1H).

Protocol of route f: $PVBLG_{70}$ (25 mg, 0.1 mmol vinyl group) and 9-BBN (1 mL, 0.5 M in THF) were mixed in dry THF (1 mL) under nitrogen. The solution was stirred at room temperature overnight. $Pd(PPh_3)_4$ (2 mg, ca. 0.0017 mmol), 4'-bromoacetophenone (100 mg, 0.5 mmol) and a $NaHCO_3$ solution (3 M, 1 mL) were added under nitrogen. The mixture was stirred in a 70 °C oil bath for 20 h. The organic phase was collected, washed by a brine solution (10 mL), dried by Na_2SO_4, filtered, and concentrated under vacuum. The resulting product was washed by ether (10 mL × 2). The residue was dried under vacuum to give $(PVBLG\text{-}g)_{70}$ (22 mg, 60 % overall yield). ^{1}H NMR ($CDCl_3$/TFA-*d* = 5/1, 500 MHz): δ7.92 ($CH_3(CO)C(\underline{CH})_2(CH)_2CCH_2CH_2$-, 2H) 7.33 ($CH_3(CO)C(CH)_2(\underline{CH})_2CCH_2CH_2$-, 2H) 7.20-7.09 (-$CO_2C_6\underline{H_4}$-, 4H), 5.12 (-$CO_2\underline{CH_2}C_6H_4$-, 2H), 4.62 (αH, 1H), 3.03-2.85 (-$C_6H_4\underline{CH_2CH_2}C_6H_4$-, 4H), 2.56 (βH, 2H), 2.17 (γH, 1H), and 1.95 (γH, 1H).

Protocol of route g: $PVBLG_{70}$ (25 mg, 0.1 mmol vinyl group) was dissolved in THF (0.5 mL) containing 0.1% I2959 in a 7-mL glass vial. The solution was exposed to UV (365 nm, 20 mW/cm^2) for 10 min till gelation was observed.

General procedure for synthesis of PZLL-*b*-PVBLG In a glovebox, Lys-NCA (61 mg, 0.2 mmol) was dissolved in dry DMF (1.0 mL). HMDS stock solution in DMF (0.1 M × 100 μL) was added to the stirred NCA solution. The reaction mixture was stirred for 16 h at room temperature and then an aliquot of the solution was used for GPC analysis. VB-Glu-NCA (145 mg, 0.5 mmol) dissolved in DMF/NB (1 mL/100 μL) was added to the rest of the PZLL polymerization solution. TBD stock solution in DMF (0.01 M × 20 μL) was then added to the mixture. The mixture was monitored by FT-IR and removed from the glovebox after VB-Glu-NCA was completely consumed. An aliquot of the polymerization solution was used for GPC analysis for the M_n and MWD of the resulting PZLL-*b*-PVBLG. The remaining solution was treated by TBAF (0.1 M, 50 μL), diisopropylethylamine (10 μL) and benzyl chloroformate

(20 μL) to protect the *N*-terminus. The solution was stirred at room temperature for 2-3 h followed by the removal of the majority of the solvent under vacuum. The residue was precipitated with ether (30 mL). The obtained PZLL-*b*-PVBLG was sonicated for 5 min in ether and centrifuged to remove the solvent. After the sonication-centrifugation procedure was repeated two more times, PZLL-*b*-PVBLG was collected and dried under vacuum (108 mg, 62 % yield) and analyzed by ^{1}H NMR. ^{1}H NMR ($CDCl_3$/TFA-*d* = 5/1, 500 MHz). PVBLG block: δ 7.38 (Ar<u>H</u>, 100H), 7.21 (Ar<u>H</u>, 100H), 6.64 (dd, J_1 =11.0 Hz, J_2 =18.0 Hz $C_6H_4C\underline{H}=CH_2$, 50H), 5.76 (d, *J* =18.0 Hz, $C_6H_4CH=C\underline{H}_2$, 50H), 5.22 (d, *J* =11.0 Hz, $C_6H_4CH=C\underline{H}_2$, 50H), 5.04-5.14 (b, Ar$\underline{CH}_2$, 100H), 4.61 (s, αH, 50H), 2.45 (s, γH, 100H), 2.14 (s, βH, 50H), 1.95 (s, βH, 50H) (*Note: all the chemical shifts of PVBLG protons in this block polymer downshifted ~0.2 ppm from the PVBLG homopolymer reported above because the two samples were using different solvents and therefore had different internal standard*). PZLL block: δ 7.38 (d, *J* =7.0 Hz, Ar<u>H</u>, 100H), 5.04-5.14 (b, Ar$\underline{CH}_2$, 40H), 4.42 (s, αH, 20H), 3,13 (s, εH, 40H), 1.63 (s, βH, 40H), 1.45-1.27 (δH and γH, 80H).

Circular dichroism (CD analysis of (PVBLG-d)$_{70}$ The CD experiments were performed on a J-720 CD spectrometer. The solution was placed in a quartz disc with a light path of 0.5 cm. The mean residue molar ellipticity was calculated based on the ellipticity obtained, concentration of polymer and the molar weight of the repeating unit ([θ] in deg·cm^2·dmol^{-1}) = (millidegrees × mean residue weight)/(pathlength in millimeters × concentration of polypeptide in mg/mL).

Brief discussion of the CD spectra of (PVBLG-d)$_{70}$ To demonstrate that (PVBLG-d)$_{70}$ does not aggregate in water, (PVBLG-d)$_{70}$ was prepared at two different concentrations and their CD spectra were collected and compared. The CD spectra of both samples showed two minima, one at 208 nm and the other at 222 nm, indicating the formation of α-helical conformations. The mean residua molar ellipticities at both 208 nm and 222 nm of these two samples were identical, suggesting that (PVBLG-d)$_{70}$ stayed monomeric at these concentrations in water.

Cytotoxicity Measurements The cytotoxicity of the polymers was characterized using the MTT cell viability assay (Sigma-Aldrich, St. Louis, MO, USA). HeLa cells were seeded in 96-wells plates at 0.5×10^4 cells/well and grown overnight at 37 °C, 5% CO_2 in medium containing 10 % horse serum and 1 % penicillin-streptomycin. The medium was replaced with serum-supplemented DMEM 24 h later and the polymer was added to the cells at known final concentrations. After incubation for 48 h, the medium was removed and the cells were washed by PBS. Reconstituted 3-[4,5-dimethylthiazol-2-yl]-2,5-diphenyl tetrazolium bromide (MTT, 10 μL) was added. The plates were then incubated for four hours and MTT solubilization solution (100 μL, Sigma-Aldrich, St. Louis, MO, USA) was added. The absorbance at 570 nm was recorded using a PerkinElmer 1420 multilable counter plate reader (Waltham, MA, USA). The background absorbance of cells killed with ethanol was subtracted from the viable cell absorbance and normalized to cells grown in DMEM (Figure S6).

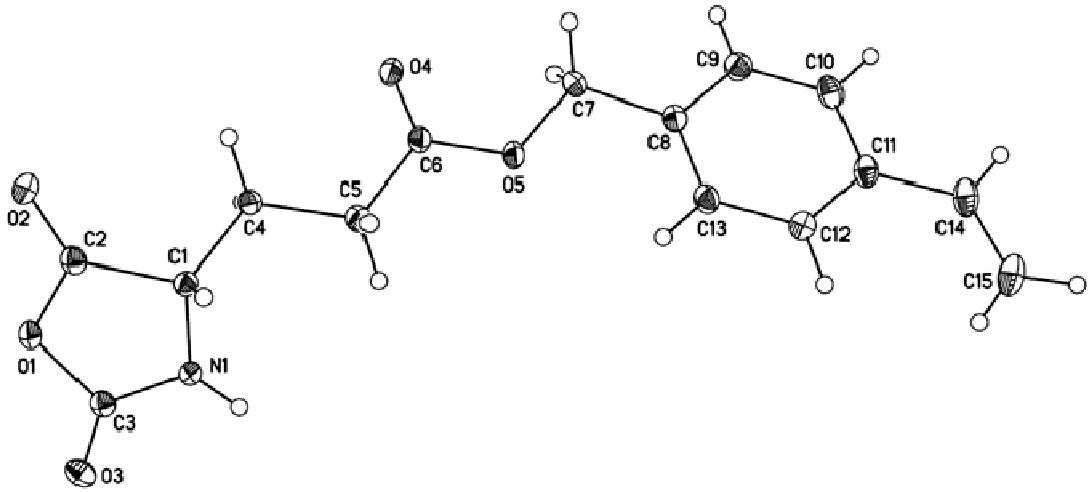
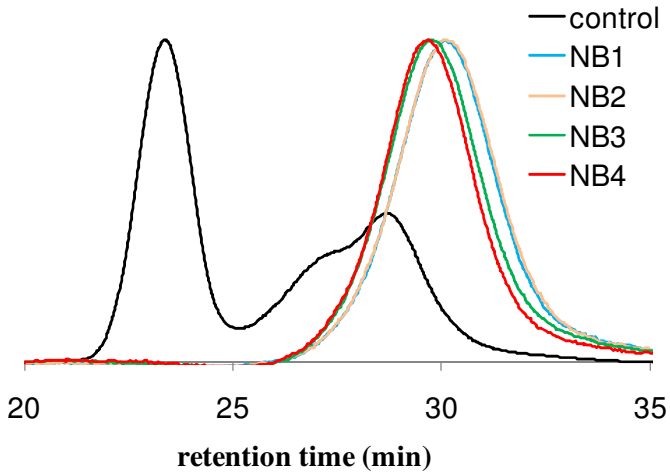
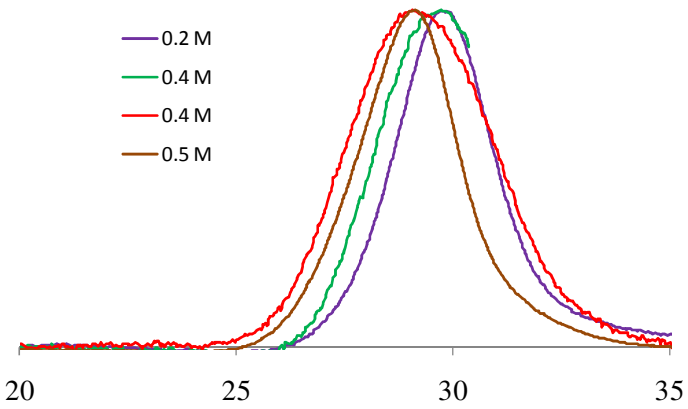


Figure S1. X-Ray diffraction image of VB-Glu-NCA. The details were given at the end of the Supporting Information (see Appendix 1 S17-S27).



	M_n ($\times 10^{-4}$)	MWD
expected	1.22	NA
control	1.87	2.03
NB1	1.21	1.13
NB2	1.11	1.17
NB3	1.43	1.10
NB4	1.41	1.07

Figure S2. The overlay of GPC (MALLS detector) curves of PVBLG derived from HMDS-mediated VB-Glu-NCA polymerizations at a M/I ratio of 50:1 in the presence of various amounts of NB (control: black, 0 μL; NB1: blue, 10 μL; NB2: orange, 20 μL; NB3: green, 30 μL; NB4: red, 40 μL). The MW and MWD of the resulting PVBLGs were summarized in the table on the right panel.



	M_n ($\times 10^{-3}$)	MWD
expected	12.2	NA
0.2 M NCA	11.2	1.17
0.3 M NCA	11.1	1.12
0.4 M NCA	12.2	1.18
0.5 M NCA	15.7	1.11

Figure S3. The overlay of the GPC (MALLS detector) curves of PVBLGs derived from HMDS-mediated VB-Glu-NCA polymerizations at different initial VB-Glu-NCA concentrations at the M/I ratio of 50/1 in the mixture of DMF/NB (v/v = 500/30). The MW and MWD of the resulting PVBLGs were summarized in the table on the right panel.

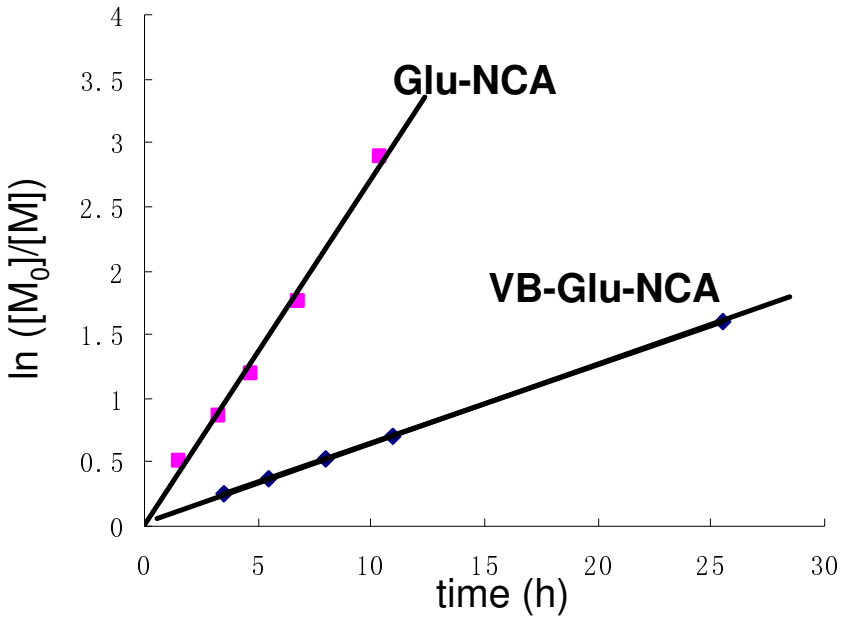


Figure S4. Kinetic study of Glu-NCA and VB-Glu-NCA polymerization in DMF at room temperature. The initial NCA concentration was 0.2 mM. The conversion of the NCA was determined by monitoring the FTIR anhydride band at 1784 cm^{-1}.

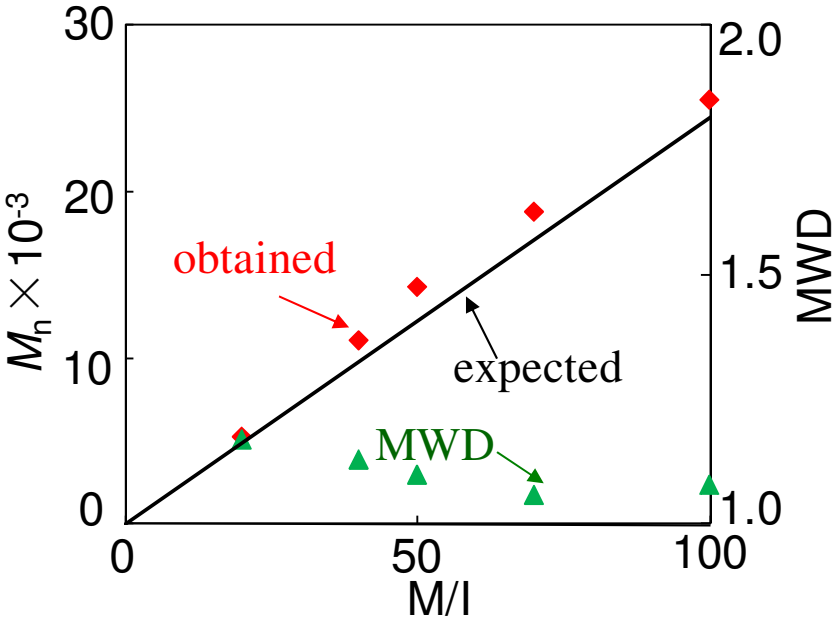


Figure S5. Plot of M_n and MWD of HMDS-mediated VB-Glu-NCA polymerization at various M/I ratios in the mixture of DMF/NB (v/v = 500/30) without co-catalyst.

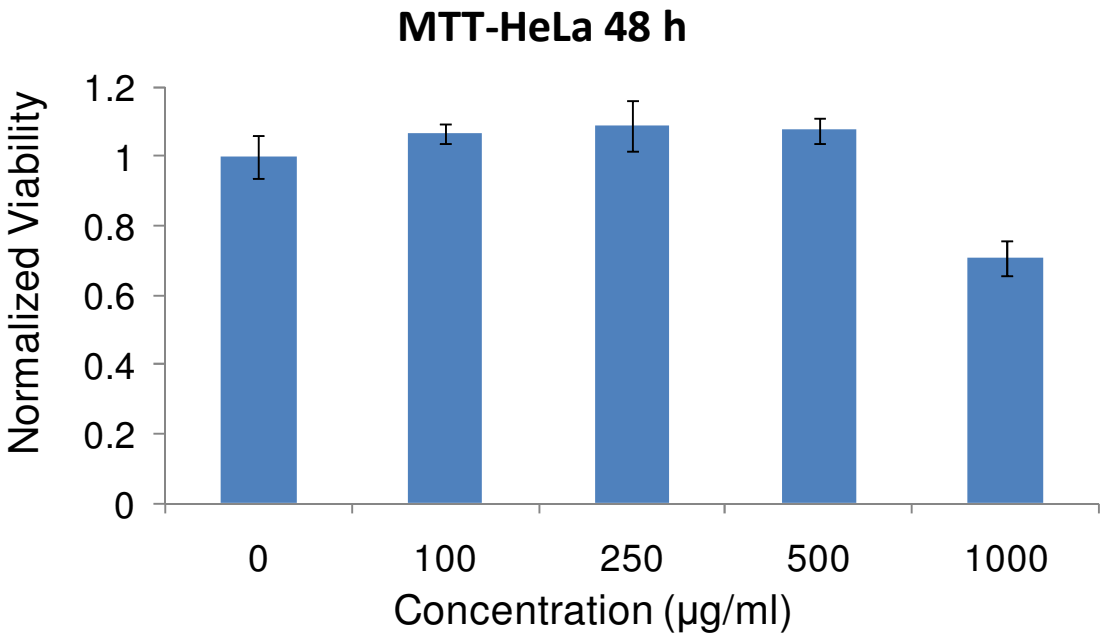


Figure S6. Cytotoxicity evaluation of $(PVBLG\text{-}d)_{70}$ using MTT assay in HeLa cells.

Reference

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Appendix 1: Details of Crystal Structure of VB-Glu-NCA (Figure S1)

Table 1. Crystal data and structure refinement for b50j2.

Identification code	b50j2	
Empirical formula	C15 H15 N O5	
Formula weight	289.28	
Temperature	100(2) K	
Wavelength	1.54178 Å	
Crystal system	Orthorhombic	
Space group	P2(1)2(1)2(1)	
Unit cell dimensions	a = 5.8000(2) Å	a= 90°.
	b = 7.4540(2) Å	b= 90°.
	c = 31.7146(11) Å	g = 90°.
Volume	1371.12(8) $Å^3$	
Z	4	
Density (calculated)	1.401 Mg/m^3	
Absorption coefficient	0.890 mm^{-1}	
F(000)	608	
Crystal size	0.393 x 0.158 x 0.063 mm^3	
Theta range for data collection	2.79 to 66.21°.	
Index ranges	-6<=h<=5, -8<=k<=8, -37<=l<=33	
Reflections collected	7817	
Independent reflections	2267 [R(int) = 0.0203]	
Completeness to theta = 66.21°	96.0 %	
Absorption correction	Semi-empirical from equivalents	
Max. and min. transmission	0.9461 and 0.7211	
Refinement method	Full-matrix least-squares on F^2	
Data / restraints / parameters	2267 / 0 / 194	
Goodness-of-fit on F^2	1.054	
Final R indices [I>2sigma(I)]	R1 = 0.0221, wR2 = 0.0558	

R indices (all data)	R1 = 0.0228, wR2 = 0.0565
Absolute structure parameter	0.11(14)
Largest diff. peak and hole	0.132 and -0.145 e.Å^{-3}

Table 2. Atomic coordinates (x 10^4) and equivalent isotropic displacement parameters ($Å^2$x 10^3) for b50j2. U(eq) is defined as one third of the trace of the orthogonalized U^{ij} tensor.

	x	y	z	U(eq)
N(1)	8138(2)	2980(1)	3010(1)	18(1)
O(1)	7474(2)	3584(1)	2335(1)	19(1)
O(2)	4365(2)	1884(1)	2208(1)	25(1)
O(3)	10560(2)	4893(1)	2647(1)	25(1)
O(4)	479(2)	2196(1)	3783(1)	25(1)
O(5)	2550(2)	713(1)	4262(1)	22(1)
C(1)	6193(2)	1842(2)	2906(1)	17(1)
C(2)	5817(2)	2366(2)	2448(1)	18(1)
C(3)	8937(2)	3910(2)	2680(1)	19(1)
C(4)	4066(2)	2146(2)	3179(1)	17(1)
C(5)	4364(2)	1276(2)	3611(1)	21(1)
C(6)	2250(2)	1463(2)	3882(1)	19(1)
C(7)	583(2)	821(2)	4541(1)	21(1)
C(8)	1293(2)	187(2)	4973(1)	20(1)
C(9)	-276(2)	394(2)	5300(1)	24(1)
C(10)	243(2)	-206(2)	5704(1)	26(1)
C(11)	2355(2)	-1007(2)	5795(1)	23(1)
C(12)	3919(2)	-1210(2)	5463(1)	23(1)
C(13)	3393(2)	-638(2)	5058(1)	22(1)
C(14)	2851(3)	-1590(2)	6230(1)	29(1)
C(15)	4789(3)	-2296(2)	6372(1)	36(1)

Table 3. Bond lengths [Å] and angles [°] for b50j2.

N(1)-C(3)	1.3371(16)
N(1)-C(1)	1.4490(16)
N(1)-H(1N)	0.834(16)
O(1)-C(2)	1.3699(15)
O(1)-C(3)	1.4065(15)
O(2)-C(2)	1.1912(15)
O(3)-C(3)	1.1976(15)
O(4)-C(6)	1.2056(16)
O(5)-C(6)	1.3422(15)
O(5)-C(7)	1.4442(14)
C(1)-C(2)	1.5183(16)
C(1)-C(4)	1.5242(16)
C(1)-H(1)	1.0000
C(4)-C(5)	1.5269(16)
C(4)-H(4A)	0.9900
C(4)-H(4B)	0.9900
C(5)-C(6)	1.5024(17)
C(5)-H(5A)	0.9900
C(5)-H(5B)	0.9900
C(7)-C(8)	1.5071(17)
C(7)-H(7A)	0.9900
C(7)-H(7B)	0.9900
C(8)-C(9)	1.3896(18)
C(8)-C(13)	1.3904(19)
C(9)-C(10)	1.3891(18)
C(9)-H(9)	0.9500
C(10)-C(11)	1.3930(19)
C(10)-H(10)	0.9500
C(11)-C(12)	1.3990(18)
C(11)-C(14)	1.4736(17)
C(12)-C(13)	1.3872(18)
C(12)-H(12)	0.9500
C(13)-H(13)	0.9500
C(14)-C(15)	1.321(2)
C(14)-H(14)	0.9500

C(15)-H(15A)	0.9500
C(15)-H(15B)	0.9500

C(3)-N(1)-C(1)	113.30(10)
C(3)-N(1)-H(1N)	122.3(11)
C(1)-N(1)-H(1N)	123.0(11)
C(2)-O(1)-C(3)	109.46(8)
C(6)-O(5)-C(7)	115.11(10)
N(1)-C(1)-C(2)	100.28(9)
N(1)-C(1)-C(4)	114.44(9)
C(2)-C(1)-C(4)	112.90(10)
N(1)-C(1)-H(1)	109.6
C(2)-C(1)-H(1)	109.6
C(4)-C(1)-H(1)	109.6
O(2)-C(2)-O(1)	121.87(10)
O(2)-C(2)-C(1)	129.42(11)
O(1)-C(2)-C(1)	108.71(10)
O(3)-C(3)-N(1)	131.12(12)
O(3)-C(3)-O(1)	120.83(11)
N(1)-C(3)-O(1)	108.05(10)
C(1)-C(4)-C(5)	110.86(10)
C(1)-C(4)-H(4A)	109.5
C(5)-C(4)-H(4A)	109.5
C(1)-C(4)-H(4B)	109.5
C(5)-C(4)-H(4B)	109.5
H(4A)-C(4)-H(4B)	108.1
C(6)-C(5)-C(4)	112.37(10)
C(6)-C(5)-H(5A)	109.1
C(4)-C(5)-H(5A)	109.1
C(6)-C(5)-H(5B)	109.1
C(4)-C(5)-H(5B)	109.1
H(5A)-C(5)-H(5B)	107.9
O(4)-C(6)-O(5)	122.25(11)
O(4)-C(6)-C(5)	126.09(11)
O(5)-C(6)-C(5)	111.66(10)
O(5)-C(7)-C(8)	108.82(10)
O(5)-C(7)-H(7A)	109.9

C(8)-C(7)-H(7A)	109.9
O(5)-C(7)-H(7B)	109.9
C(8)-C(7)-H(7B)	109.9
H(7A)-C(7)-H(7B)	108.3
C(9)-C(8)-C(13)	118.56(11)
C(9)-C(8)-C(7)	117.78(12)
C(13)-C(8)-C(7)	123.64(11)
C(10)-C(9)-C(8)	120.72(13)
C(10)-C(9)-H(9)	119.6
C(8)-C(9)-H(9)	119.6
C(9)-C(10)-C(11)	121.33(12)
C(9)-C(10)-H(10)	119.3
C(11)-C(10)-H(10)	119.3
C(10)-C(11)-C(12)	117.40(11)
C(10)-C(11)-C(14)	119.48(12)
C(12)-C(11)-C(14)	123.12(12)
C(13)-C(12)-C(11)	121.44(13)
C(13)-C(12)-H(12)	119.3
C(11)-C(12)-H(12)	119.3
C(12)-C(13)-C(8)	120.54(12)
C(12)-C(13)-H(13)	119.7
C(8)-C(13)-H(13)	119.7
C(15)-C(14)-C(11)	127.16(13)
C(15)-C(14)-H(14)	116.4
C(11)-C(14)-H(14)	116.4
C(14)-C(15)-H(15A)	120.0
C(14)-C(15)-H(15B)	120.0
H(15A)-C(15)-H(15B)	120.0

Symmetry transformations used to generate equivalent atoms:

Table 4. Anisotropic displacement parameters ($Å^2$x 10^3) for b50j2. The anisotropic displacement factor exponent takes the form: $-2p^2[h^2 a^{*2}U^{11} + ... + 2 h k a^* b^* U^{12}]$

	U^{11}	U^{22}	U^{33}	U^{23}	U^{13}	U^{12}
N(1)	16(1)	21(1)	15(1)	1(1)	-1(1)	-2(1)
O(1)	22(1)	20(1)	16(1)	2(1)	2(1)	0(1)
O(2)	25(1)	31(1)	17(1)	-1(1)	-3(1)	-1(1)
O(3)	22(1)	25(1)	28(1)	2(1)	5(1)	-5(1)
O(4)	20(1)	37(1)	18(1)	4(1)	0(1)	5(1)
O(5)	20(1)	30(1)	15(1)	4(1)	2(1)	3(1)
C(1)	17(1)	16(1)	16(1)	0(1)	0(1)	0(1)
C(2)	19(1)	17(1)	17(1)	-1(1)	3(1)	4(1)
C(3)	19(1)	18(1)	19(1)	-1(1)	2(1)	3(1)
C(4)	17(1)	18(1)	16(1)	-1(1)	1(1)	0(1)
C(5)	19(1)	26(1)	18(1)	2(1)	2(1)	2(1)
C(6)	21(1)	21(1)	16(1)	0(1)	0(1)	-2(1)
C(7)	18(1)	27(1)	18(1)	2(1)	3(1)	0(1)
C(8)	23(1)	19(1)	18(1)	0(1)	0(1)	-3(1)
C(9)	23(1)	28(1)	22(1)	1(1)	2(1)	3(1)
C(10)	32(1)	28(1)	19(1)	-1(1)	8(1)	1(1)
C(11)	32(1)	19(1)	18(1)	0(1)	0(1)	-2(1)
C(12)	24(1)	23(1)	22(1)	0(1)	-1(1)	0(1)
C(13)	24(1)	26(1)	17(1)	0(1)	4(1)	1(1)
C(14)	45(1)	24(1)	17(1)	-1(1)	3(1)	0(1)
C(15)	51(1)	35(1)	21(1)	3(1)	-6(1)	2(1)

Table 5. Hydrogen coordinates (x 10^4) and isotropic displacement parameters ($Å^2$x 10^3) for b50j2.

	x	y	z	U(eq)
H(1N)	8870(30)	2890(20)	3235(5)	28(4)
H(1)	6667	554	2922	20
H(4A)	2697	1630	3037	21
H(4B)	3806	3450	3214	21
H(5A)	4717	-14	3574	25
H(5B)	5689	1838	3757	25
H(7A)	-683	61	4432	25
H(7B)	25	2074	4556	25
H(9)	-1721	951	5248	29
H(10)	-864	-68	5922	31
H(12)	5373	-1753	5515	28
H(13)	4475	-810	4837	27
H(14)	1644	-1443	6429	35
H(15A)	6053	-2474	6187	43
H(15B)	4918	-2625	6661	43

Table 6. Torsion angles [°] for b50j2.

C(3)-N(1)-C(1)-C(2)	-4.29(13)
C(3)-N(1)-C(1)-C(4)	-125.41(11)
C(3)-O(1)-C(2)-O(2)	179.57(11)
C(3)-O(1)-C(2)-C(1)	0.29(12)
N(1)-C(1)-C(2)-O(2)	-176.96(12)
C(4)-C(1)-C(2)-O(2)	-54.74(16)
N(1)-C(1)-C(2)-O(1)	2.25(11)
C(4)-C(1)-C(2)-O(1)	124.47(10)
C(1)-N(1)-C(3)-O(3)	-175.93(12)
C(1)-N(1)-C(3)-O(1)	4.74(14)
C(2)-O(1)-C(3)-O(3)	177.58(11)
C(2)-O(1)-C(3)-N(1)	-3.01(12)
N(1)-C(1)-C(4)-C(5)	-77.02(13)
C(2)-C(1)-C(4)-C(5)	169.11(10)
C(1)-C(4)-C(5)-C(6)	-176.96(10)
C(7)-O(5)-C(6)-O(4)	1.06(17)
C(7)-O(5)-C(6)-C(5)	-179.44(10)
C(4)-C(5)-C(6)-O(4)	0.29(18)
C(4)-C(5)-C(6)-O(5)	-179.19(10)
C(6)-O(5)-C(7)-C(8)	-171.37(10)
O(5)-C(7)-C(8)-C(9)	172.25(10)
O(5)-C(7)-C(8)-C(13)	-9.81(17)
C(13)-C(8)-C(9)-C(10)	0.2(2)
C(7)-C(8)-C(9)-C(10)	178.22(12)
C(8)-C(9)-C(10)-C(11)	1.0(2)
C(9)-C(10)-C(11)-C(12)	-1.0(2)
C(9)-C(10)-C(11)-C(14)	178.76(13)
C(10)-C(11)-C(12)-C(13)	0.05(19)
C(14)-C(11)-C(12)-C(13)	-179.75(12)
C(11)-C(12)-C(13)-C(8)	1.1(2)
C(9)-C(8)-C(13)-C(12)	-1.16(19)
C(7)-C(8)-C(13)-C(12)	-179.08(12)
C(10)-C(11)-C(14)-C(15)	-176.88(14)
C(12)-C(11)-C(14)-C(15)	2.9(2)

Symmetry transformations used to generate equivalent atoms:

Table 7. Hydrogen bonds for b50j2 [Å and °].

D-H...A	d(D-H)	d(H...A)	d(D...A)	<(DHA)
N(1)-H(1N)...O(4)#1	0.834(16)	2.039(16)	2.8628(13)	169.4(14)

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