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ATRP from an Amino Acid-Based Initiator: A Facile Approach for α-Functionalized Polymers

Shrinivas Venkataraman and Karen L. Wooley*

Center for Materials Innovation and Department of Chemistry, Washington University in Saint Louis, One Brookings Drive, Campus Box 1134, Saint Louis, Missouri 63130

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Introduction

The development of living radical polymerization (LRP) techniques constitutes one of the key developments in the field of synthetic polymer chemistry, allowing for the synthesis of a variety of polymers with low molecular weight distributions and well-defined architectures, with precise control over the compositions and structures.[1−5] Atom transfer radical polymerization (ATRP) has emerged as a versatile technique that has been applied broadly because of the robustness of the chemistry and the commercial availability of many initiators, catalysts, and ligands. As with other LRP techniques, control of the α-chain terminus is accessed from the initiator design, which for ATRP most often involves an alkyl halide with an activating substituent on the α-carbon that undergoes homolytic cleavage in the presence of catalyst to initiate polymerization. Since ester groups are good activating groups, α-haloester-based compounds are commonly used as ATRP initiators, whereby the ester unit can also carry functionality. The functionality incorporated into the polymer via an ester linkage is prone to hydrolysis; however, this lability has been overcome by their replacement with amide linkages through the use of α-halo amide-based initiators.[6,7]

The presence of amides is often considered to present unique challenges for ATRP in the production of polymers of narrow molecular weight distribution and having molecular weights in agreement with theoretical values. A recent report from Haddleton's laboratory thoroughly examined the conditions under which amide containing ATRP initiators gave well-controlled polymerization of various methacrylates and styrene.[7] Prior work in Sawamoto's laboratory demonstrated the preparation of poly(methyl methacrylate) with a narrow polydispersity index (PDI) using *N*,*N*-dimethyl-2-bromopropanamide as the initiator.[6] Matyjaszewski's laboratory has studied the polymerization of methacrylamides using model α-haloamide-based initiators, obtaining well-defined block copolymers.[8] Recently, Xia et al. have employed various chloropropionamides as initiators to polymerize *N*-isopropylacrylamide with narrow molecular weight distributions to study the influence of the end group composition resulting from the initiators on the thermal properties.[9]

Chain end composition in amphiphilic block copolymers can be utilized to prepare functional nanostructures with regiochemical control.[10,11−13] In our laboratory, we have prepared nanostructures originating from a functional initiator, which upon polymerization and followed by post-polymerization reactions and self-assembly, will project amino acids and other biologically active ligands from their surfaces.[14−19] In one synthetic strategy, the growth of block copolymers of *tert*-butyl acrylate and styrene from α-bromo butyryl amido-terminated peptides loaded upon a solid support was followed by cleavage from the resin and deprotection to release an amphiphilic triblock of peptide, poly(acrylic acid), and poly(styrene).[20] Determination of the polymerization behavior was complicated by the attachment of the polymer chain to the resin during growth and the unusual solubility and electrolyte character of the amphiphilic structure after cleavage. Herein, therefore, we discuss results from studies that investigated the employment of an amino acid-based ATRP initiator to polymerize *tert*-butyl acrylate and styrene in solution.

Experimental Procedures

Materials. All chemicals, solvents, and reagents were purchased from Sigma Aldrich (Saint Louis, MO) or Fisher Scientific (Pittsburgh, PA). CH_2Cl_2 (99.9%), triethylamine (TEA) (99.9%), *tert*-butyl acrylate (*t*BA) (98%), and styrene (99%) were distilled over CaH_2 and stored under N_2. $SOCl_2$ (>99%) was doubly distilled before use. CuBr (99.999%), $CuBr_2$ (99.999%), *p*-dimethoxy benzene (*p*-DMB) (99%), *N*,*N*-dimethyl formamide (DMF, 99.8%), PMDETA, (99%), methanol (99.93%), L-valine, and 2-bromopropionyl bromide (97%) were used as received.

Methods. ^1H NMR spectra were recorded at 300 MHz on a Varian Mercury 300 spectrometer on solutions in $CDCl_3$ or CD_2Cl_2 with the solvent proton signal as a standard. ^{13}C NMR spectra were recorded at 75.4 MHz in $CDCl_3$ on a Varian Mercury 300 spectrometer with the solvent carbon signal as a standard. IR spectra were obtained on a Perkin-Elmer BX FT-IR system equipped with a diffuse reflectance accessory. Glass transition temperatures (T_g) were measured by differential scanning calorimetry (DSC) at a heating rate of 10 °C/min on a Mettler Toledo DSC822e using Mettler Toledo Star SW 7.01 software. T_g values were taken at the midpoint of the inflection tangent on the third heating scan. T_m values were taken at the onset of the transition.

Size-exclusion chromatography (SEC) was conducted on a Waters 1515 HPLC (Waters Chromatography, Inc., Milford, MA) equipped with a Waters 2414 differential refractometer, a PD2026 dual-angle (15 and 90°) light-scattering detector (Precision Detectors, Inc., Franklin, MA), and three-column series PL gel 5 μm Mixed C, 500 Å, and 10^4 Å, 300 mm × 7.5 mm columns (Polymer Laboratories Inc., Amherst, MA). The system was equilibrated at 35 °C in THF, which served as the polymer solvent and eluent with a flow rate of 1.0 mL/min. Polymer solutions were prepared at a known concentration (ca. 3 mg/mL), and an injection volume of 200 μL was used. Data collection and analysis were performed, respectively, with Precision Acquire software and Discovery 32 software (Precision Detectors, Inc., Franklin, MA). Interdetector delay volume and the light-scattering detector calibration constant were determined by calibration using a nearly monodispersed polystyrene standard (M_p = 90 kDa, M_w/M_n < 1.04, Pressure Chemical Co., Pittsburgh, PA). The differential refractometer was calibrated with a standard polystyrene reference material (SRM 706 NIST) of known S-5 specific refractive index increment d*n*/d*c* (0.184 mL/g). The d*n*/d*c* values of the analyzed polymers were then determined from the differential refractometer response.

Synthesis of Methyl 2-Amino-3-methylbutanoate·HCl (2).[21] L-valine (0.99 g, 8.5 mmol, 1.0 equiv) was suspended in 40 mL of methanol in a 100 mL two-necked round-bottomed flask fitted with a N_2 inlet and rubber septa. The suspension was cooled in an ice bath, and $SOCl_2$ (4.7 mL, 7.7 g, 65 mmol, 7.6 equiv) was added dropwise. The suspension became a homogeneous solution. Deionized water (0.6 mL, 0.6 g, 30 mmol, 4 equiv) was then added, and the reaction mixture was allowed to stir for 24 h at room temperature. The solvent was removed in vacuo, and the resultant solid was washed twice with methanol (2 mL) to result in a white solid, which was recrystallized from acetone to yield white needle-like crystals. Yield: 0.96 g (68%). T_m = 163 °C. IR: 3462, 3009,

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* Corresponding author. Tel.: (314) 935-7136; fax: (314) 935-9844; e-mail: klwooley@wustl.edu.

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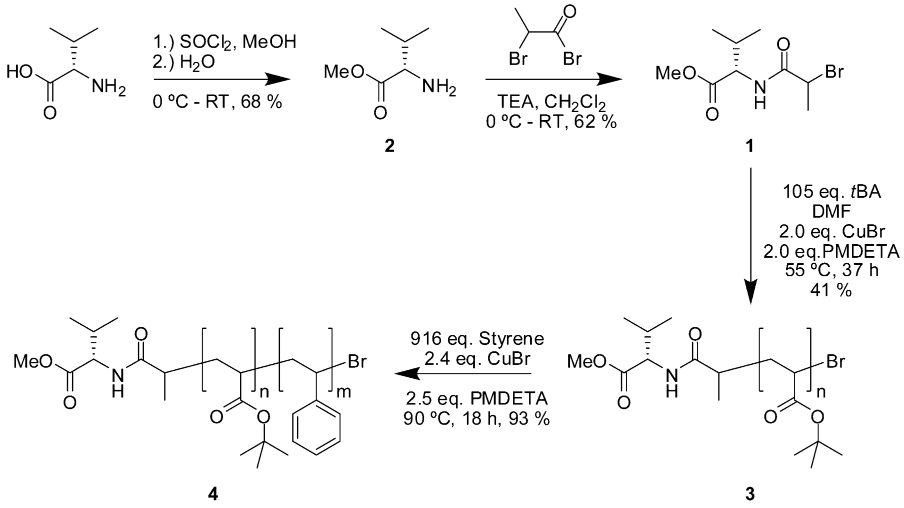
Scheme 1. Synthesis of P*t*BA-*b*-PS (4) Diblock Copolymer from L-Valine-Based ATRP Initiator (1)



2824, 2617, 1982, 1759, 1504, 1287, 1240 cm^{-1}. ^{1}H NMR (CDCl$_3$ δ, ppm): 8.87 (br, s, N***H***$_2$), 3.94 (m, C***H***CO), 3.83 (s, OC$\boldsymbol{H}_3$), 2.47 (m, C***H***(CH$_3$)$_2$), 1.15 (m, CH(C$\boldsymbol{H}_3$)$_2$) ^{13}C NMR (CDCl$_3$ δ, ppm): 168.98, 58.65, 53.20, 30.17, 18.57.

Synthesis of Methyl 2-[(2-Bromopropanoyl)amino]-3-methylbutanoate (1). Into a 250 mL two-necked round-bottomed flask fitted with a N$_2$ inlet and a rubber septum was placed **2** (1.0 g, 7.4 mmol, 1.0 equiv), which was allowed to dissolve in CH$_2$Cl$_2$ (20 mL) and then was cooled in an ice bath. TEA (2.0 mL, 1.5 g, 15 mmol, 2 equiv) and 2-bromopropionyl bromide (0.85 mL, 1.7 g, 8.1 mmol, 1.1 equiv) were added dropwise via gastight syringes, and the reaction was allowed to proceed at room temperature overnight. The reaction mixture was then washed with deionized water (3 × 100 mL), dried over MgSO$_4$, and concentrated in vacuo to afford the crude product as a dark colored oil. The product (R_f = 0.65) was purified by silica flash column chromatography using CH$_3$OH/CH$_2$Cl$_2$ (1:9) as an eluent to yield 1.22 g (62%) as an orange oil. IR: 3312, 2960−2870, 1754, 1693, 1681, 1538, 1469, 1440, 1374, 1311, 1270, 1213, 1150, 988, 761 cm^{-1}. ^{1}H NMR (CDCl$_3$, δ, ppm): 4.52 (m, COC***H***NH), 4.44 (m, C***H***Br), 3.76 (s, OC$\boldsymbol{H}_3$), 2.25 (m, C***H***(CH$_3$)$_2$), 1.88, 1.89 (m, CH(C$\boldsymbol{H}_3$)Br), 0.95 (m, CH(C$\boldsymbol{H}_3$)$_2$). ^{13}C NMR (CDCl$_3$ δ, ppm): 172.16, 172.28, 169.49, 169.53, 57.70, 57.91, 52.56, 44.69, 45.21, 31.52, 31.74, 23.10, 23.34, 17.85, 17.95, 19.14. MS: theor m/z = 288.0211 [M + Na]$^+$, exptl m/z = 288.0221 [M + Na]$^+$

Preparation of P*t*BA Macroinitiator (3). Into a 100 mL Schlenk flask, **1** (0.41 g, 1.5 mmol, 1.0 equiv), DMF (35.0 mL), and *t*-BA (25.0 mL, 20.8 g, 162 mmol, 105 equiv) were placed, after a freeze−pump−thaw cycle, CuBr (0.45 g, 3.1 mmol, 2.0 equiv) was added, and again a freeze−pump−thaw cycle was performed. PMDETA (0.55 g, 3.2 mmol, 2.0 equiv) was added, and the reaction mixture was degassed by performing three freeze−pump−thaw cycles and allowing the reaction mixture to equilibrate to room temperature. The reaction mixture was immersed into an oil bath at 55 °C. Aliquots were removed via N$_2$ washed gastight syringes, and the reaction was monitored by ^{1}H NMR spectroscopy and SEC. The reaction was quenched at 37 h by immersing in a liquid N$_2$ bath. The reaction mixture was dissolved in ∼200 mL of THF, and the copper catalyst was removed by passing through a neutral alumina column. The solution was concentrated and precipitated thrice in an ice cold 60:40 methanol/water mixture to yield P*t*BA as a white powder. Yield = 3.14 g (41%, based on conversion). M_n^{NMR} = 8.6 kDa, M_n^{GPC} = 8.3 kDa, and PDI = 1.11. T_g = 41 °C. IR: 3462, 3433, 2977, 1716, 1446, 1366, 1251, 1142, 844, 751 cm^{-1}. ^{1}H NMR (CDCl$_3$, δ, ppm): 4.50 (C***H***Br end group), 4.10 (COC***H***NH end group), 3.73 (OC$\boldsymbol{H}_3$, end group), 2.10−2.39 (br, C***H*** of the polymer backbone), 1.68−2.00 (br, meso C$\boldsymbol{H}_2$ of the polymer backbone), 1.24−1.65 (br, meso and racemo C$\boldsymbol{H}_2$ of the polymer backbone), 1.20−1.50 (br, (C$\boldsymbol{H}_3$)$_3$C), 0.95 (CH(C$\boldsymbol{H}_3$)$_2$ end group) ^{13}C NMR (CDCl$_3$, δ, ppm): 174.4, 80.6, 42.6, 42.2, 37.7, 36.1, 28.3.

Preparation of P*t*BA-*b*-PS Block Copolymer (4). In a 50 mL Schlenk flask, the P*t*BA macroinitiator, (M_n^{GPC} = 8.3 kDa, PDI = 1.11, 1.01 g, 0.11 mmol, 1.0 equiv), CuBr (38 mg, 0.26 mmol, 2.4 equiv), and styrene (11.6 mL, 10.5 g, 101 mmol, 916 equiv) were mixed. A freeze−pump−thaw cycle was conducted followed by the addition of PMDETA (60 μL, 50 mg, 0.28 mmol, 2.5 equiv). The reaction mixture was degassed by performing three freeze−pump−thaw cycles and then was immersed into an oil bath maintained at 90 °C. The reaction was allowed to proceed for 18 h and was quenched by immersing in a liquid N$_2$ bath. The reaction mixture was dissolved in THF (50 mL), and the copper catalyst was removed by passing the solution through a neutral alumina column. The polymer solution was concentrated and was precipitated thrice in 50:50 methanol/water mixtures to result in the block copolymer. Yield = 1.8 g (93%, based on conversion). M_n^{NMR} = 20.2 kDa, M_n^{GPC} = 22.5 kDa, and PDI = 1.22. $(T_g)_{\mathrm{P}t\mathrm{BA}}$ = 51 °C and $(T_g)_{\mathrm{PS}}$ = 97 °C. IR: 3434, 3024, 1977, 2922, 1943, 1871, 1803, 1724, 1601, 1494, 1452, 1367, 1254, 1144, 847, 752, 697 cm^{-1}. ^{1}H NMR (CD$_2$Cl$_2$, δ, ppm): 6.90−7.45 (br, ortho- and para-Ar***H***), 6.30−6.90 (br, meta-Ar***H***), 4.40 (C***H***Br end group), 4.10 (COC***H***-NH end group), 3.72 (OC$\boldsymbol{H}_3$, end group), 1.72−2.07 (br, meso C$\boldsymbol{H}_2$ of the polymer backbone) 2.04−2.40 (br, C***H*** of the polymer backbone), 1.20−1.50 (br, (C$\boldsymbol{H}_3$)$_3$C), 1.24−1.65 (br, meso and racemo C$\boldsymbol{H}_2$ of the polymer backbone), 0.94 (CH(C$\boldsymbol{H}_3$)$_2$ end group). ^{13}C NMR (CD$_2$Cl$_2$, δ, ppm): 174.6, 146.0, 128.6, 126.2, 80.9, 40.1−46.5, 38.0, 36.5, 30.7, 28.5.

Kinetics of *t*-BA Polymerization using (1) as Initiator. Into an oven-dried 10 mL Schlenk flask was placed **1** (106.2 mg, 0.40 mmol, 1.0 equiv), CuBr (56.2 mg, 0.40 mmol, 1.0 equiv), CuBr$_2$ (2.5 mg, ca. 5% relative to CuBr), *t*-BA (4.98 g 39 mmol, 97 equiv), and *p*-DMB (100.0 mg ca. 2% relative to monomer as an internal ^{1}H NMR standard to estimate monomer conversion), and the reaction mixture was degassed by performing a freeze−pump−thaw cycle. PMDETA (0.8 μL, 0.4 mmol, 1.0 equiv) was added via a gastight syringe, and the reaction mixture was degassed by performing three freeze−pump−thaw cycles, and upon equilibration to room temperature after the third cycle, the flask was immersed into an oil bath pre-maintained at 60 °C. Aliquots (ca. 0.2 μL) were removed via a N$_2$ washed gastight syringe. ^{1}H NMR spectroscopy was used to determine the monomer conversion. SEC samples were prepared by dissolving the crude mixture into THF, and the resultant solution was passed through a short column of neutral alumina to remove the copper catalyst.

Results and Discussion

Amphiphilic polymers originating from a functional initiator offer, upon self-assembly in water, regiochemical control of the

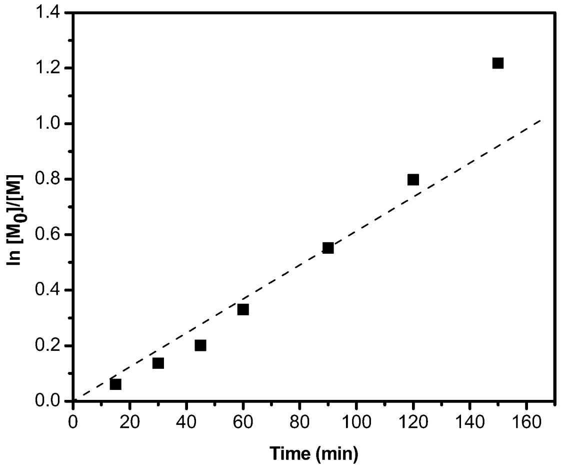


Figure 1. First-order kinetic plot for the bulk polymerization of *t*BA at 60 °C, $\mathbf{1}_0/[CuBr]_0/[CuBr_2]_0/[PMDETA]_0/[tBA]_0 = 1:1:0.05:1:100$.

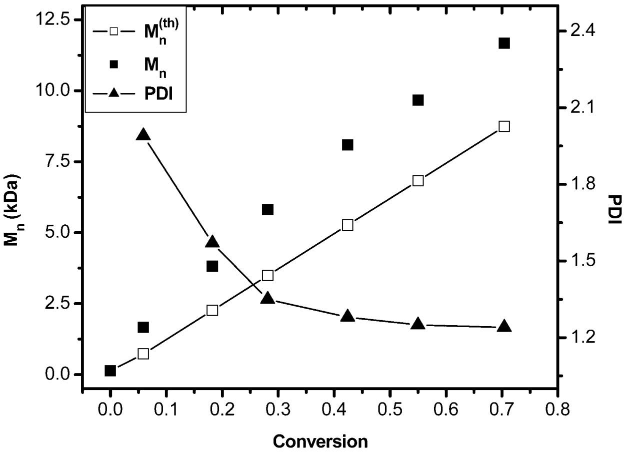


Figure 2. Evolution of M_n and PDI with conversion for the polymerization of *t*BA in the bulk at 60 °C, $\mathbf{1}_0/[CuBr]_0/[CuBr_2]_0/[PMDETA]_0/[tBA]_0 = 1:1:0.05:1:100$; open squares with line, theoretical M_n; closed squares, experimental M_n; and closed triangles, PDI.

surface and/or core compositions of the resultant nanostructures by the design of the polymer sequence. In our study, we chose amino acids as the precursor for the preparation of functional initiators due to their biological relevance, commercial availability, amphiphilic nature, and simplicity.

Valine was chosen as a model amino acid, and it was readily converted into an ATRP initiator, **1**. To avoid potential problems with the carboxylic acid functionality, such as poor control over the polymerization due to the interference of the acid with the catalyst system, as reported previously,[22] the free carboxylic acid was esterified using methanol in the presence of thionyl chloride.[21] Amidation of the amino group of the methyl ester of L-valine, **2**, with 2-bromopropionyl bromide in the presence of TEA afforded the desired initiator **1** (Scheme 1), which was purified by silica flash column chromatography eluting with CH_3OH/CH_2Cl_2 (1:9).

Kinetic studies of the polymerization of *t*BA, initiated by **1**, were conducted at 60 °C in the bulk, employing CuBr and PMDETA as the catalyst system. To impart better control over the polymerization, 5% $CuBr_2$, relative to CuBr, was added.[23] The first-order kinetic plot of polymerization of *t*BA illustrates that the polymerization occurred in a controlled manner and that the monomer consumption increased linearly with time (Figure 1). The linearity also suggests that there is a constant radical concentration throughout the polymerization.

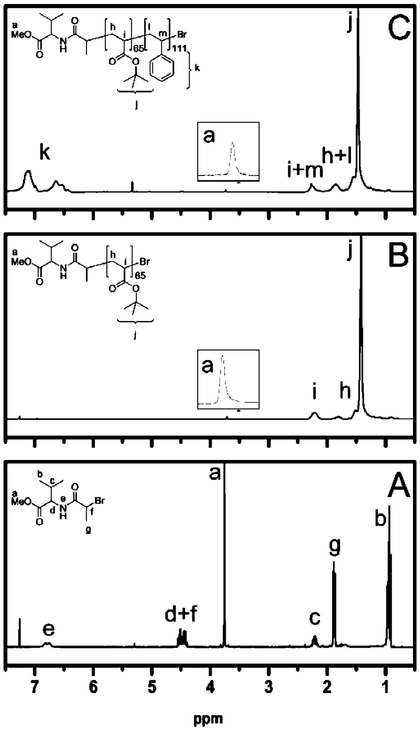


Figure 3. ^1H NMR spectra (300 MHz): (A) initiator **1** (in $CDCl_3$); (B) P*t*BA macroinitiator **3** (in $CDCl_3$), with inset (3.8−3.65 ppm) showing the methoxy chain end of the polymer; (C) P*t*BA-*b*-PS diblock copolymer **4** (in CD_2Cl_2), with inset (3.8−3.65 ppm) showing the methoxy chain end of the polymer.

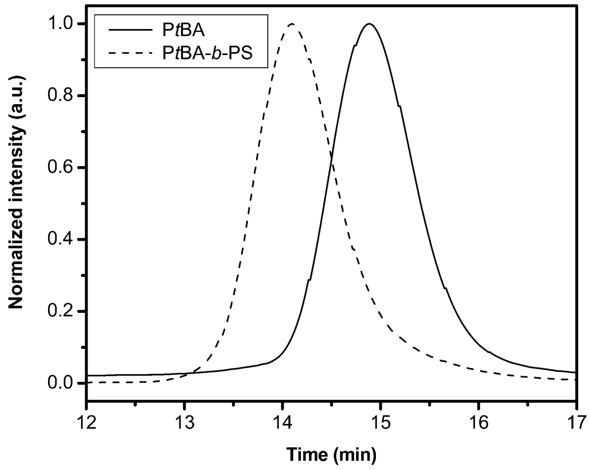


Figure 4. SEC traces of P*t*BA macroinitiator and P*t*BA-*b*-PS block copolymer prepared from **1**.

Evolution of M_n with conversion was found to be linear as shown in Figure 2 (squares). As observed previously,[7] experimental M_n values were found to be higher than the theoretical values throughout the polymerization. This phenomenon could be partially attributed to the fact that the experimental M_n values are obtained from PS calibration, which does not account for the difference in the polymer−column interaction for P*t*BA. The PDI was high (~2) initially and decreased with conversion, a behavior observed commonly under living radical polymerization conditions. The final PDI at 70% conversion was found to be 1.25 (Figure 2 (triangles)).

To demonstrate the living characteristic of this polymerization system, a P*t*BA macroinitiator prepared from **1** was used for the preparation of a block copolymer. The P*t*BA employed was found have $M_n^{GPC} = 8300$ Da (PDI = 1.11), and this was in good agreement with the $M_n^{NMR} = 8600$ Da, which was determined by comparing the resonances of methoxy proton from the chain end and the backbone C*H* resonances. This P*t*BA was chain extended with styrene, and the resultant block copolymer, P*t*BA-*b*-PS, was found to have $M_n^{GPC} = 22\,500$ Da and PDI = 1.22. Figure 3B,C illustrates the ^{1}H NMR spectra of the macroinitiator and the block copolymer, respectively. The aromatic resonances from 6.5−7.2 ppm show that the polymerization of styrene occurred. The inset shows the resonances corresponding to the methoxy protons from the initiator, demonstrating that the initiator functionalities are retained for both the P*t*BA macroinitiator and the block copolymer.

Overlaid SEC traces corresponding to the macroinitiator and P*t*BA-*b*-PS diblock copolymer are shown in Figure 4. The shift in peak elution time to the left indicates that the chain extension has occurred.

Conclusion

A methyl ester-protected ATRP initiator from L-valine was demonstrated to provide for the initiation and controlled polymerization of *t*BA, using commercially available ligands, to afford well-defined polymers that retained the valine functionality at the α-chain terminus. Control during the polymerizations was confirmed by kinetic studies of the *t*BA polymerization and by extension of the resulting macroinitiator into a diblock copolymer with the polymerization of styrene. Each macroinitiator and block copolymer had a narrow PDI but still experienced the problem of the experimental molecular weight being in excess of the theoretical value. Nonetheless, the ability to prepare well-defined block copolymers will allow for their conversion into amino-acid labeled nanostructures, for which this study can be extended potentially to other amino acids and peptides. This work also complements the highly interesting recent examples of protein−polymer conjugates or soft hybrid materials, involving the growth of polymers directly from proteins in a controlled fashion.[12,24,25]

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References and Notes

(1) Matyjaszewski, K.; Xia, J. *Chem. Rev.* **2001**, *101*, 2921−2990.

(2) Kamigaito, M.; Ando, T.; Sawamoto, M. *Chem. Rev.* **2001**, *101*, 3689−3746.

(3) Hawker, C. J.; Bosman, A. W.; Harth, E. *Chem. Rev.* **2001**, *101*, 3661−3688.

(4) Chiefari, J.; Chong, Y. K.; Ercole, F.; Krstina, J.; Jeffery, J.; Le, T. P. T.; Mayadunne, R. T. A.; Meijs, G. F.; Moad, C. L.; Moad, G.; Rizzardo, E.; Thang, S. H. *Macromolecules* **1998**, *31*, 5559−5562.

(5) Perrier, S.; Takolpuckdee, P. *J. Polym. Sci., Part A: Polym. Chem.* **2005**, *43*, 5347−5393.

(6) Baek, K.-Y.; Kamigaito, M.; Sawamoto, M. *J. Polym. Sci., Part A: Polym. Chem.* **2002**, *40*, 1937−1944.

(7) Limer, A.; Haddleton, D. M. *Macromolecules* **2006**, *39*, 1353−1358.

(8) Teodorescu, M.; Matyjaszewski, K. *Macromolecules* **1999**, *32*, 4826−4831.

(9) Xia, Y.; Burke, N. A. D.; Stover, H. D. H. *Macromolecules* **2006**, *39*, 2275−2283.

(10) Hawker, C. J.; Wooley, K. L. *Science* **2005**, *309*, 1200−1205.

(11) Klok, H.-A.; Becker, S.; Schuch, F.; Pakula, T.; Müllen, K. *Macromol. Chem. Phys.* **2002**, *203*, 1106−1113.

(12) Bontempo, D.; Maynard, H. D. *J. Am. Chem. Soc.* **2005**, *127*, 6508−6509.

(13) Reynhout, I. C.; Löwik, D. W. P. M.; van Hest, J. C. M.; Cornelissen, J. J. L. M.; Nolte, R. J. M. *Chem. Commun.* **2005**, 602−604.

(14) Joralemon, M. J.; Smith, N. L.; Holowka, D.; Baird, B.; Wooley, K. L. *Bioconjugate Chem.* **2005**, *16*, 1246−1256.

(15) Cheng, C.; Khoshdel, E.; Wooley, K. L. *Macromolecules* **2005**, *38*, 9455−9465.

(16) Qi, K.; Ma, Q.; Remsen, E. E.; Clark, C. G.; Wooley, K. L. *J. Am. Chem. Soc.* **2004**, *126*, 6599−6607.

(17) Joralemon, M. J.; Murthy, K. S.; Remsen, E. E.; Becker, M. L.; Wooley, K. L. *Biomacromolecules* **2004**, *5*, 903−913.

(18) Becker, M. L.; Remsen, E. E.; Pan, D.; Wooley, K. L. *Bioconjugate Chem.* **2004**, *15*, 699−709.

(19) Wooley, K. L. *J. Polym. Sci., Part A: Polym. Chem.* **2000**, *38*, 1397−1407.

(20) Becker, M. L.; Liu, J.; Wooley, K. L. *Biomacromolecules* **2005**, *6*, 220−228.

(21) Mustafi, D.; Sachleben, J. R.; Wells, G. B.; Makinen, M. W. *J. Am. Chem. Soc.* **1990**, *112*, 2558−2566.

(22) Zhang, X.; Matyjaszewski, K. *Macromolecules* **1999**, *32*, 7349−7353.

(23) Matyjaszewski, K.; Patten, T. E.; Xia, J. *J. Am. Chem. Soc.* **1997**, *119*, 674−680.

(24) Klok, H.-A. *J. Polym. Sci., Part A: Polym. Chem.* **2005**, *43*, 1−17.

(25) Lele, B. S.; Murata, H.; Matyjaszewski, K.; Russell, A. J. *Biomacromolecules* **2005**, *6*, 3380−3387.

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