Synthesis of Carboxy-Functionalized Heterobifunctional Poly(ethylene glycol) by a Thiol-Anionic Polymerization Method

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Received April 4, 2006; Revised Manuscript Received July 6, 2006

ABSTRACT: Potassium *n*-butyl 3-thiolate-1-propionate (K−nBMP) and dipotassium 3-thiolate-1-propionate (K−MPA) initiated anionic polymerization of ethylene oxide (EO) to yield carboxy-functionalized poly(ethylene glycol)s (HOOC−PEG) with predicted number-average molecular weights (M_n up to 18 700) and narrow molecular weight distributions ($M_w/M_n \leq 1.25$, M_w is weight-average molecular weight). Additional thiol-containing anionic initiators such as mercaptoacetic acid and 11-mercaptoundecanoic acid resulted in PEGs having bimodal molecular weight distributions. Evaluation of a series of carboxy-functionalized oxy(gen) anionic initiators revealed that only potassium 12-hydroxydodecanoate (K−HDA) allowed for controlled polymerization of EO and yielded polymers with a narrow molecular weight distribution. The efficiency of the anionic initiators is related to their nucleophilicity. The carboxy-functionalized PEGs were also employed as a macroinitiator for synthesis of α-carboxy−PEG-*block*-poly(ϵ-caprolactone) copolymers.

Introduction

Poly(ethylene glycol) (PEG) has emerged as the gold standard hydrophilic polymer of choice for use in pharmaceutical, biotechnical, and biomedical applications.[1,2] The unique and versatile properties of PEG include high solubility in aqueous media and various organic solvents as well as established in vivo biocompatibility, nontoxicity (mol wt > 1000) and nonbiodegradability.[1] Recently, the conjugation of PEG, a process referred to as PEGylation, to small molecules and proteins, as well as colloidal particles and devices has been used as a means to improve or alter the properties of these substrates. For example, peptides, proteins, and small molecules have been PEGylated to formulate or deliver these therapeutic molecules.[1,3−5] The attachment of PEG to the surface of colloidal drug carriers, such as liposomes, has provided a way to increase the in vivo circulation lifetime of these vehicles.[6] In the field of biomedical engineering, the construction of tethered PEG brushes on surfaces has become a main strategy for reducing the nonspecific interactions of proteins and cells with biomedical devices.[7] Furthermore, a PEG layer may be utilized as a platform to enable molecular recognition and sensing if the free end of the tethered PEG chains are available for further conjugation to specific ligands.[8]

PEG may be directly attached to substrates, through reaction with the terminal hydroxyl groups; however, most often conjugation to the substrate first requires the activation of PEG by the addition of specific functional groups at one or both terminals of the polymer chain. In general, PEG may be synthesized by the anionic ring-opening polymerization of EO monomer.[1] Therefore, functional groups may be introduced by use of specific initiation or termination agents or the PEG chain may be modified by post-polymerization.[9,10] One of the most commonly used functionalized derivatives of PEG is methoxy-terminated PEG (MePEG) which is often employed for preparation of protein, peptide and drug conjugates.[11] MePEG−polymer conjugates and MePEG−lipid also serve as the building blocks for preparation of drug delivery vehicles.[6,12−14] The presence of the nonreactive methoxyl group at one terminus of the polymer chain allows for the homogeneous preparation of conjugates, in the absence of cross-linking reactions.[11] For specific applications, it is desirable to use heterobifunctional PEGs with distinct reactive functional groups at the ends of the polymer chain.[11,15] Heterobifunctional PEGs are required for use in targeted drug delivery and biosensor applications.

The synthesis of heterobifunctional PEGs with controlled molecular structures and high functionality is particularly challenging. The preferred functional groups that are commonly introduced onto PEG due to requirements for specific applications include amino, carboxylic acid, aldehyde, and pyridyl disulfide.[9,16−19] To date, only a few groups have reported the preparation of heterobifunctional PEGs that include a carboxylic acid group at one chain end. Riener et al. prepared α-amino-ω-carboxylic acid−PEG via a post-polymerization procedure that involved several synthetic and purification steps.[20] Ishii et al. reported on the synthesis of α-pyridyl disulfide-ω-carboxy−PEG by anionic polymerization using potassium allyl alkoxide as the initiator with conversion of the ω-end alkoxide group to a carboxylate group using succinic anhydride as the termination agent.[21] The α-allyl group was then modified to a thioester group, using thioacetic acid and azoisobutyronitrile, with selective hydrolysis to produce the 2-pyridyldithio group at the α-end using *n*-propylamine in the presence of 2,2′-dithiopyridine. However, the use of succinic anhydride to produce the carboxy-terminated PEG results in an ester linkage between the PEG chain and the succinate group. Ester linkages have limited stability in the biological milieu as they are susceptible to hydrolytic cleavage in aqueous media.[1,8] Zhang et al. reported synthesis of heterobifunctional PEG containing terminal amino and carboxylate groups by use of (cyanomethyl)potassium as the protected initiator. Following polymerization, the terminal hydroxyl group was converted to an amino group and the cyano group was hydrolyzed to a carboxylate group using an alkaline solution.[22]

To date, synthesis of carboxy-terminated PEG through use of an initiator with an unprotected functional group for polymerization has not been reported. Recently, our laboratory evaluated a range of protected and unprotected initiators for

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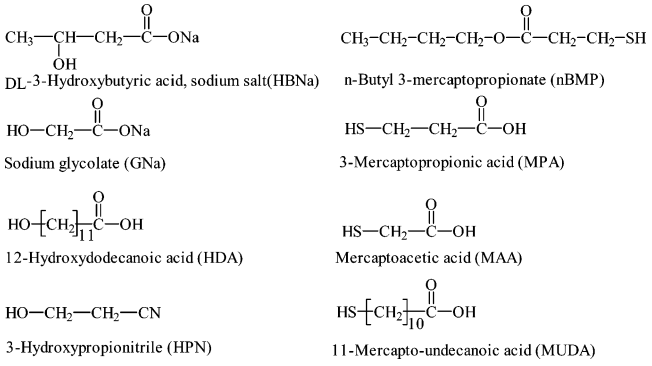
10.1021/ma0607665 CCC: $33.50
Published on Web 08/26/2006

Scheme 1. Initiators Used for the Polymerization of Ethylene Oxide in Tetrahydrofuran at 40 °C



synthesis of carboxy-functionalized heterobifunctional PEGs. It was found that a potassium thiolate effectively initiates the polymerization of EO to yield α-carboxy-*ω*-hydroxy PEGs.

This paper reports the synthesis of heterofunctionalized PEG with a carboxyl group at one chain end (α-terminal) and a hydroxyl group at the other chain end (*ω*-terminal) using thiol-anionic functional initiators (potassium thiolates). Specifically, various oxy-anionic and thiol-anionic functional initiators containing a protected or an unprotected carboxylic acid group, as shown in Scheme 1, were used to propagate the polymerization of EO. The successful synthesis of the α-carboxy-*ω*-hydroxy PEG was further demonstrated by use of this material as a macroinitiator for preparation of a series of α-carboxy−PEG-*block*-poly(*ϵ*-caprolactone) (i.e., COOH−PEG-*b*-PCL) copolymers. In addition, α-carboxy-*ω*-methoxy PEG was also prepared by termination of the polymerization with methyl iodide (MeI). Overall, this report describes a straightforward and direct method for the synthesis of heterobifunctional PEGs containing a carboxylic acid functional group at one chain end for use in a wide range of applications.

Experimental Section

Materials. Ethylene oxide (99.5%, Sigma-Aldrich) was condensed and dried by *n*-butyllithium at −78 °C. *ϵ*-Caprolactone (CL, 99%) from Aldrich was dried by calcium hydride and distilled before use. Mercaptoacetic acid (MAA, 97%), 3-mercaptopropionic acid (MPA, 99%) and *n*-butyl 3-mercaptopropionate (nBMP, 98%), all from Sigma-Aldrich, were dried by molecular sieves. 11-mercaptoundecanoic acid (MUDA, 95%) was dried under vacuum for 48 h at room temperature. Tetrahydrofuran (THF) was refluxed over fresh sodium-benzophenone complex and was distilled under a nitrogen atmosphere prior to use. Sodium glycolate (GNa, 97%, Alfa Aesar), 12-hydroxydodecanoic acid (HDA, 97%), 3-hydroxypropionitrile (HPN, 99%), sodium salt of DL-3-hydroxybutyric acid (HBNa, 99%, ACROS Organics), HCl (1.6 M in diethyl ether, HCl ether), sodium (99.5%), potassium (99.95%), naphthalene (98%), and all other chemicals purchased from Sigma-Aldrich Inc. were used as received.

Synthesis of α-Carboxy-*ω*-Hydroxy PEG Using Oxy(gen)-Anionic Polymerization. Synthesis of the carboxy-terminated PEG was conducted using a high vacuum line. The oxy-anions shown in Scheme 1 were tried as initiators for EO polymerization. A typical procedure was as follows: THF (100 mL) and HBNa (0.212 g, 2.0 mmol) were added to a flame-dried flask and the solution was cooled to 0 °C. Potassium naphthalene (2.0 mL, 1.0 M, 2.0 mmol) was then added dropwise to the solution. The reaction was maintained for 1.0 h during which time the solution remained light green in color which indicated the presence of a slight excess of potassium naphthalene. EO (10.0 g, 227.2 mmol) was then transferred to the flask at 0 °C using the capillary technique and the reactor was sealed. The reaction was maintained for 48 h at 40 °C prior to termination by the addition of 0.4 mL of HCl (36%). The potassium chloride precipitate was removed by filtration. The final product was recovered by precipitation in diethyl ether. The product was dried under vacuum for 24 h at room temperature. The monomer conversion was determined gravimetrically. Yield 2.0 g (20% by weight, Table 1, entry 1). M_n = 2600 by gel permeation chromatography (GPC) and M_w/M_n = 1.42. ^{1}H NMR ($CDCl_3$, 400 MHz): 1.17 ppm (HOOC−CH_2−CH(***CH***$_3$)−O−), 2.31 ppm (HOOC−***CH***$_2$−CH(CH_3)−O−), and 3.63 ppm (−***CH***$_2$−CH_2−O and HOOC−CH_2−***CH***(CH_3)−O−).

Synthesis of α-Carboxy-*ω*-Hydroxy PEG Using Thiol-Anionic Polymerization. Similarly, the carboxy-functionalized thiol initiators shown in Scheme 1 were tried as initiators for EO polymerization. In a typical experiment, the nBMP (0.41 g, 2.5 mmol) and THF (50 mL) were added to a flame-dried round-bottom flask. Potassium−naphthalene (2.5 mL, 1.0 M, 2.5 mmol) was added to the reaction solution dropwise at 0 °C until a pale green color appeared, which is an indication of consumption of nBMP. EO (5.0 g, 5.7 mL, 2.5 mmol) was then transferred to the reactor by the capillary technique. The reactor was sealed and maintained for 48 h at 40 °C prior to termination by the addition of 0.4 mL of HCl (36%). The potassium chloride precipitate was removed by filtration. The final product was recovered by precipitation in diethyl ether. The product was dried under vacuum for 24 h at room temperature. The monomer conversion was determined gravimetrically. Yield: 4.9 g (98% by weight, Table 2, entry 1). M_n = 1200 by GPC and M_w/M_n = 1.25. ^{1}H NMR ($CDCl_3$, 400 MHz): 0.91 ppm (***CH***$_3$−CH_2CH_2−CH_2−O), 1.38 ppm (CH_3−***CH***$_2$−CH_2−CH_2−O), 1.55 pm (CH_3−CH_2−***CH***$_2$−CH_2−O), 2.78 ppm (CO-***CH$_2$CH$_2$***−S−***CH$_2$***) and 3.63 ppm (−***CH***$_2$−CH_2−O and CH_3−CH_2−CH_2−***CH***$_2$−O−).

Synthesis of α-Carboxy-*ω*-Methoxy PEG Using Anionic Polymerization. For the synthesis of α-carboxy-*ω*-methoxy−PEG (HOOC−PEG−Me) a procedure similar to that described above was employed. However, once the monomer was consumed, a stoichiometric amount of methyl iodide (MeI) was added and the termination reaction was allowed to proceed for 24 h at room temperature. The potassium iodide (KI) was removed by filtration and the polymer was recovered by precipitation in diethyl ether. The polymer was characterized by GPC and ^{1}H NMR microscopy analysis. ^{1}H NMR ($CDCl_3$, 400 MHz, Table 3, entry 3a): 2.78 ppm (CO−***CH$_2$CH$_2$***−S−***CH$_2$***), 3.38 ppm (***CH***$_3$−O−) and 3.63 ppm (−***CH***$_2$−CH_2−O).

Table 1. Polymerization of Ethylene Oxide Using Carboxy-Functionalized Oxy-Anionic Initiators in Tetrahydrofuran at 40 °C ($[M]_0$ = 2.27 M)

entry	Init[a]	$[EO]_0/[Init]_0$	time (h)	convn[b] (%)	$M_{n,cal}$[c]	$M_{n,GPC}$[d]	M_w/M_n[e]	f[g]
1	HBNa	113	48	20	5000	2600	1.42	0.52
2	HBNa	227	24	22	10 000	1426	1.49	0.142
3	HBNa	454	48	25	20 000	9700	1.50	0.48
4	GNa	113	48	0	5000			0
5	HDA	45	40	90	2000	3400	1.11	1.0
6	HPN	113	18	50	5000	2980	1.38	0.59
7	HPN	113	40	48	5000	2900	1.58	0.58
8	HPN	227	24	35	10 000	8700	1.27	0.87

[a] Abbreviations for initiators (Init) refer to those in Scheme 1. [b] Polymer yield (defined here as monomer conversion (Conv.)) was measured gravimetrically as described in methods section. [c] $M_{n,cal}$ = 44$[EO]_0/[Init]_0$, calculated molecular weight. [d] Molecular weight measured by gel permeation chromatography. [e] Molecular weight distribution = (weight-average molecular weight)/(number-average molecular-weight) as determined by gel permeation chromatography. [g] Initiator efficiency = $M_{n,GPC}/M_{n,cal}$.

Table 2. Polymerization of Ethylene Oxide Using Carboxy-Functionalized Thiol-Anionic Initiators in Tetrahydrofuran at 40 °C ($[M]_0 = 2.27$ M)

run	Init[a]	$[EO]_0/[Init]_0$	time (h)	convn[b] (wt %)	$M_{n,cal}$[c]	$M_{n,NMR}$[d]	$M_{n,GPC}$[e]	M_w/M_n[f]	f[g]
1	nBMP	23	24	98	1000	950	1200	1.25	0.95
2	nBMP	45	24	95	2000	1920	2700	1.24	0.91
3	nBMP	113	48	92	5000	5100	7200	1.18	1.0
4	MUDA	113	24	98	5000	6300	9820	1.19	1.0
5	MUDA	227	24	97	10 000	9600	13 600	1.20	0.6
6	MAA	113	24	85	5000		4700	1.30	0.94
7	MAA	113	24	87	5000		7300	1.56	1.0
8	MAA	113	24	87	5000		5800	1.21	1.0
9	MPA	113	48	96	5000	5100	7300	1.07	1.0

[a] Abbreviations for initiators (Init) refer to those in Scheme 1. [b] Polymer yield (defined here as monomer conversion) was measured gravimetrically as described in methods section. [c] $M_{n,cal} = 44[EO]_0/[Init]_0$, calculated molecular weight. [d] Molecular weight measured by ^{1}H NMR spectroscopy. [e] Molecular weight measured by gel permeation chromatography. [f] Molecular weight distribution = (weight-average molecular weight)/(number-average molecular-weight) as determined by gel permeation chromatography. [g] Initiator efficiency = $M_{n,GPC}/M_{n,cal}$.

Table 3. Synthesis of Carboxy-Functionalized Heterobifunctional Poly(ethylene glycol) Using Dipotassium 3-Mercaptopropionate as Initiator in Tetrahydrofuran at 40 °C ($[M]_0 = 2.27$ M)

entry	$[EO]_0/[Init]_0$	time (h)	convn[a]	$M_{n,cal}$[b]	$M_{n,NMR}$[c]	$M_{n,GPC}$[d]	M_w/M_n[e]	f[f]
1	23	24	98	1000	980	1300	1.15	0.98
2	45	24	98	2000	1950	2760	1.15	0.97
3	113	48	95	5000	5100	7300	1.07	1.0
3a	113	48	95	5000	5100	7190	1.09	1.0
4	227	48	75	10 000	7200	10 300	1.08	0.72
5	227	60	89	10 000	9800	14 000	1.10	0.98
6	454	70	93	20 000	18700	24 700	1.09	0.93

[a] Polymer yield (defined here as monomer conversion) was measured gravimetrically as described in methods section. [b] $M_{n,cal} = 44[EO]_0/[Init]_0$, calculated molecular weight. [c] Molecular weight measured by ^{1}H NMR spectroscopy. [d] Molecular weight measured by gel permeation chromatography. [e] Molecular weight distribution = (weight-average molecular weight)/(number-average molecular weight) as determined by gel permeation chromatography. [f] Initiator efficiency = $M_{n,GPC}/M_{n,cal}$.

Table 4. Synthesis of Carboxy-Terminated Poly(ethylene glycol)-*block*-poly(ϵ-caprolactone) Using Carboxy-Terminated Poly(ethylene glycol) as Macroinitiator in Dichloromethane for 24 h at 25 °C ($[OH]_0/[HCl]_0 = 1/3.0$, $[M]_0 = 1.46$ M)

entry	macroinitiator $M_{n,PEG}$	f_{CL}[a] (wt %)	$[M]_0/[OH]_0$	F_{CL}[b] (wt %)	$M_{n,cal((PCL)}$[c]	$M_{n,NMR(PCL)}$[d]	$M_{n,GPC}$[e]	M_w/M_n[f]
1	2000	0.50	17.5	0.49	2000	2000	7600	1.16
2	2000	0.67	35	0.65	4000	3700	11 000	1.16
3	5000	0.5	44	0.48	5000	4600	16 600	1.09
4	5000	0.67	88	0.64	10000	9000	24 400	1.18

[a] Feed composition of ϵ-caprolactone. [b] Copolymer composition calculated from ^{1}H NMR spectroscopy. [c] Theoretical molecular weight of poly(ϵ-caprolactone) (PCL) block based on feed composition. [d] Molecular weight of the PCL block as determined from the copolymer composition and the known molecular weight, M_n, of PEG. [e] Molecular weight measured by gel permeation chromatography. [f] Molecular weight distribution from gel permeation chromatography analysis.

Synthesis of HOOC−PEG-*b*-PCL. The HOOC−PEG-*b*-PCL copolymers were synthesized using a method outlined in detail elsewhere with slight modifications.[24] HOOC−PEG (2.0 g, Table 4, entry 3) was added to a flame-dried flask and dried twice by the azeotropic distillation of toluene. Dichloromethane (10 mL) and CL (2.0 g, 17.54 mmol, distilled over calcium hydride) were then added prior to the addition of 1.2 mL of the initiator, HCl (1 M, 1.2 mmol) in diethyl ether. The reaction was maintained at room temperature for 24 h prior to termination by addition of 1 mL of triethylamine. The reaction mixture was then filtered and the block copolymer was isolated by precipitation in ether. A typical ^{1}H NMR spectrum is included in the results and discussion section. The molecular weight of the PCL block was calculated from the overall composition obtained from ^{1}H NMR spectroscopy and the known molecular weight of the HOOC−PEG macroinitiator. Yield: 98% (3.9 g, Table 4, entry 2). ^{1}H NMR ($CDCl_3$, 400 MHz): 1.35 ppm (m, 2H, CO−CH_2−CH_2−***CH***$_2$−CH_2−CH_2−O), 1.55 ppm (m, 4H, CO−CH_2−***CH***$_2$−CH_2−***CH***$_2$−CH_2−O), 2.28 ppm (m, 2H, CO−***CH***$_2$−CH_2−CH_2−CH_2−CH_2−O), 2.76 ppm (6H, −***CH***$_2$−S-***CH***$_2$−CH_2−COOH), 3.63 ppm (−***CH***$_2$−CH_2−O), and 4.07 ppm (m, 2H, CO−CH_2−CH_2−CH_2−CH_2−***CH***$_2$−O).

Characterization of Polymers. ^{1}H NMR spectra were obtained on a Mercury 400 spectrometer (400 MHz for ^{1}H) in $CDCl_3$ solvent. Chemical shifts were reported in ppm with $CHCl_3$ as the internal standard. GPC measurements were carried out at room temperature using a Waters 590 liquid chromatography system equipped with three Waters Styragel HR 4E columns (50−10^4 Å pore size, 5 μm particle size) and a 410 differential refractometer detector. THF with 1% triethylamine was used as the solvent at a flow rate of 1.0 mL/min at 40 °C. Narrow polystyrene standards (Polysciences Inc., Warrington, PA) were used to generate a calibration curve. The data obtained were recorded and manipulated using the Windows-based Millenium 2.0 software package (Waters Inc., Milford, MA).

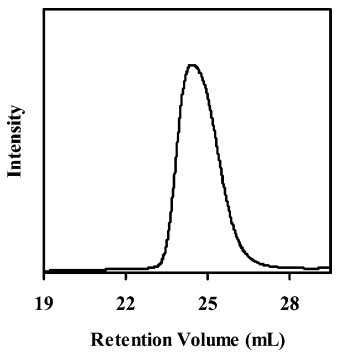


Figure 1. Gel permeation chromatography trace of the crude product for ethylene oxide polymerization initiated by potassium *n*-butyl 3-thiolate-1-propionate in tetrahydrofuran at 40 °C for 24 h: $[EO]_0 = 2.27$ M, $M_n = 2700$, $M_w/M_n = 1.24$ (Table 2, entry 2). M_n = number-average molecular weight; M_w = weight-average molecular weight; M_w/M_n = molecular weight distribution.

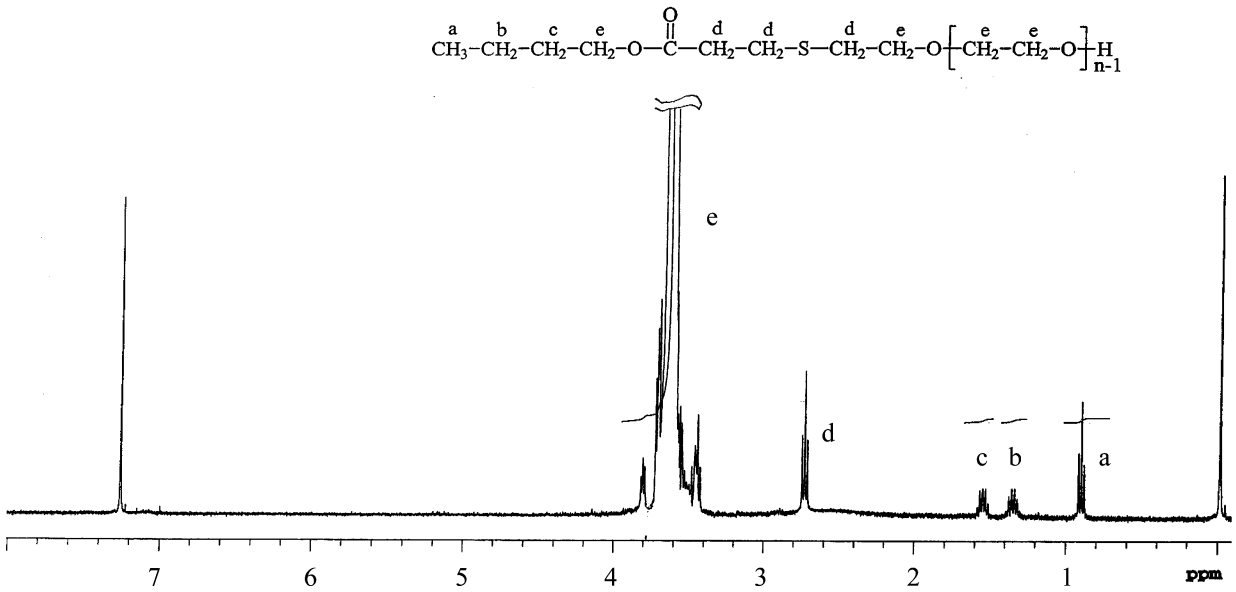


Figure 2. [1]H NMR spectrum of the *n*-butyl-3-mercaptopropionate-terminated poly(ethylene glycol) (Table 2, entry 2).

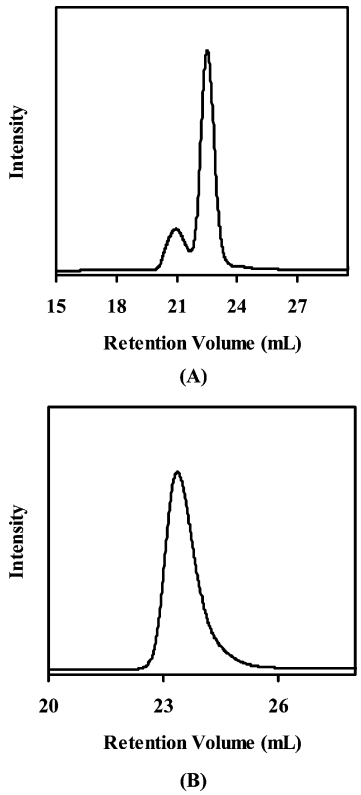


Figure 3. Gel permeation chromatography traces of mercaptoacetic acid-terminated poly(ethylene glycol) and 3-mercaptopropionic acid-terminated poly(ethylene glycol): (A) M_n = 13 600, M_w/M_n = 1.20 (Table 2, entry 5; (B) M_n = 7300, M_w/M_n = 1.07 (Table 2, entry 9). M_n = number-average molecular weight; M_w = weight-average molecular weight; M_w/M_n = molecular weight distribution.

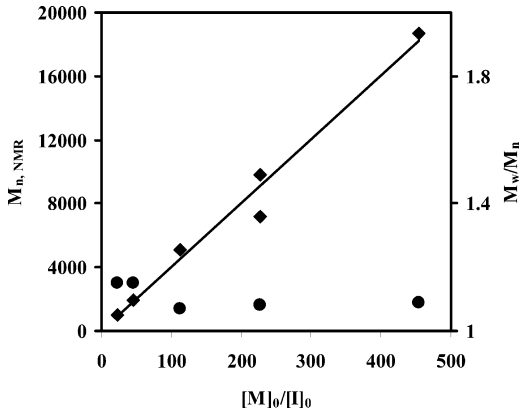
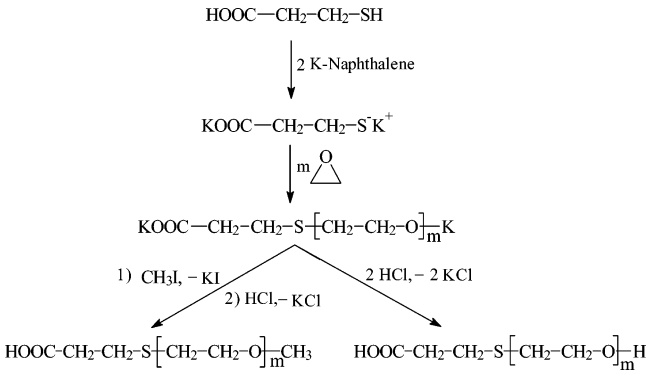


Figure 4. Dependence of molecular weight ($M_{n,NMR}$) and molecular weight distribution (M_w/M_n) on the feed ratio of monomer to initiator ($[M]_0/[I]_0 = [EO]_0/[MPA]_0$): (a) Table 3, entry 1; (b) Table 3, entry 2; (c) Table 3, entry 3; (d) Table 3, entry 5; (e) Table 3, entry 6. $M_{n,NMR}$ = number-average molecular weight as measured by [1]H NMR analysis; M_w/M_n = molecular weight distribution.

Scheme 2. Typical Synthetic Procedure for Polymerization of Poly(ethylene glycol) Using the Thiol-Anionic Polymerization Method in Tetrahydrofuran at 40 °C



Results and Discussion

Functionalized PEGs may be prepared by use of specific initiation or termination agents during the polymerization procedure or via a post-polymerization modification method.[8,10] The advantage of preparing the functionalized PEGs using the initiation method is that each polymer chain will contain exactly one of the desired functional groups. Protected functionalized initiators have been employed for anionic polymerization to produce PEG derivatives containing a carboxyl group at one chain end.[18] In our research, protected and unprotected carboxy-containing compounds with hydroxyl or thiol functional groups were used as initiators for anionic polymerization of the EO monomer. As outlined in Scheme 2, the protected or unprotected initiators with hydroxyl or thiol (mercapto) functional groups were first reacted with potassium naphthalene to yield the

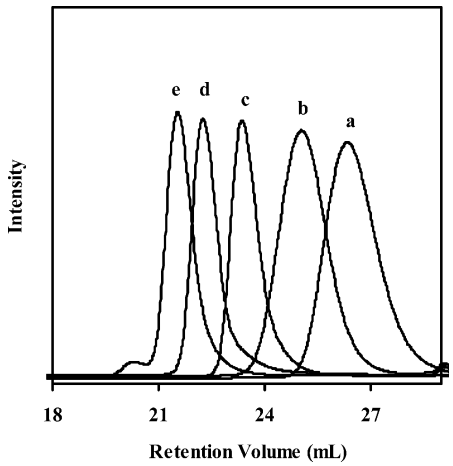


Figure 5. Molecular weight development for the EO polymerization using dipotassium 3-mercaptopropionate as initiator in tetrahydrofuran at 40 °C.

corresponding potassium anionic initiators, which resulted in a highly heterogeneous solution in THF.

1. Synthesis of α-Carboxy-ω-Hydroxy-PEG Using Oxy-Anionic Functional Initiators. As summarized in Table 1 (entries 1−8), a series of unprotected initiators including HBNa, GNa, HDA and HPN were evaluated for the oxy-anionic polymerization of EO. The potassium alkoxide initiators were prepared in situ by reaction of the carboxy-containing alcohol, as shown in Scheme 1, with potassium naphthalene (Scheme 2). The nucleophilicity of the resulting initiators increase in the following order K−HDA < K−HBNa < K−GNa.[2] Interestingly, when K−GNa was used as the initiator, no PEG polymer was obtained. Use of K−HBNa as the initiator resulted in a monomer conversion of 25% as determined by gravimetric analysis by precipitation in diethyl ether following a 48 h reaction period at 40 °C. The resulting polymer had a lower molecular weight than predicted from the monomer/initiator ratio and the molecular weight distribution of the polymer was broad (M_w/M_n > 1.40, Table 1, entries 1−3). The monomer conversion did not increase with an increase in the reaction time which indicates that the reaction was terminated during the 48 h period. In addition, GPC analysis revealed a bimodal distribution for the polymers formed using K−HBNa as the initiator. This may be attributed to the higher nucleophilicity of K−HBNa compared to K−HDA, which in turn results in an increase in the probability of side reactions. The use of K−HDA as the initiator allowed for a high monomer conversion of approximately 90%, with the initiator efficiency reaching 1, and polymer with a narrow molecular weight distribution (M_w/M_n = 1.1, Table 1, entry 5) following a 40 h reaction period. Therefore, a decrease in the nucleophilicity of the functional initiator resulted in an increase in both the extent of monomer conversion and the efficiency of the initiator. Therefore, K−HDA was the most suitable initiator for the anionic polymerization of EO. However, the drawback of the resulting polymer is the presence of the rather large hydrophobic headgroup (i.e., HOOC−$(CH_2)_{11}$−PEG), which alters the typical physical and chemical properties of HOOC−PEG. K−HPN was also an effective initiator for EO polymerization in THF; however, the resulting polymers had slightly broad molecular weight distributions (Table 1, entries 6−8). In addition, conversion of the nitrile group to a carboxyl group was unsuccessful using hydrolysis in a 45% (wt) sodium hydroxide solution at 80 °C for 4 h.[22] It has been established that scission of the main chain takes place during the alkaline hydrolysis of cyano-terminated PEG.[8] To further examine the influence of the nucleophilicity of the initiator on EO polymerization a group of carboxy-functionalized thiol initiators were selected for further evaluation.

2. Synthesis of α-Carboxy-ω-Hydroxy PEG Using Thiol-Anionic Functional Initiators. Recently, Dufresne et al. reported the synthesis of thiol-functionalized PEG using a thiol anionic polymerization method. For this process, potassium *tert*-butyl mercaptan (thiolate) was used as initiator for EO polymerization and the polymer was employed to prepare the hydrophilic block copolymers of PEG and *N,N′*-dimethylaminoethyl methacrylate (DMAEMA).[23] The functionality of the resulting polymers was reported to be 23% with an incomplete monomer conversion of 50%. To our knowledge, there is no other report on the use of thiol anionic initiators for polymerization of EO.

The nucleophilicity of an initiator must be high enough to allow for fast initiation of the monomer; yet, if the nucleophilicity of the initiator is too high this will increase the likelihood of side reactions. The potassium thiolate anions are less nucleophilic than the corresponding potassium oxygen anions.

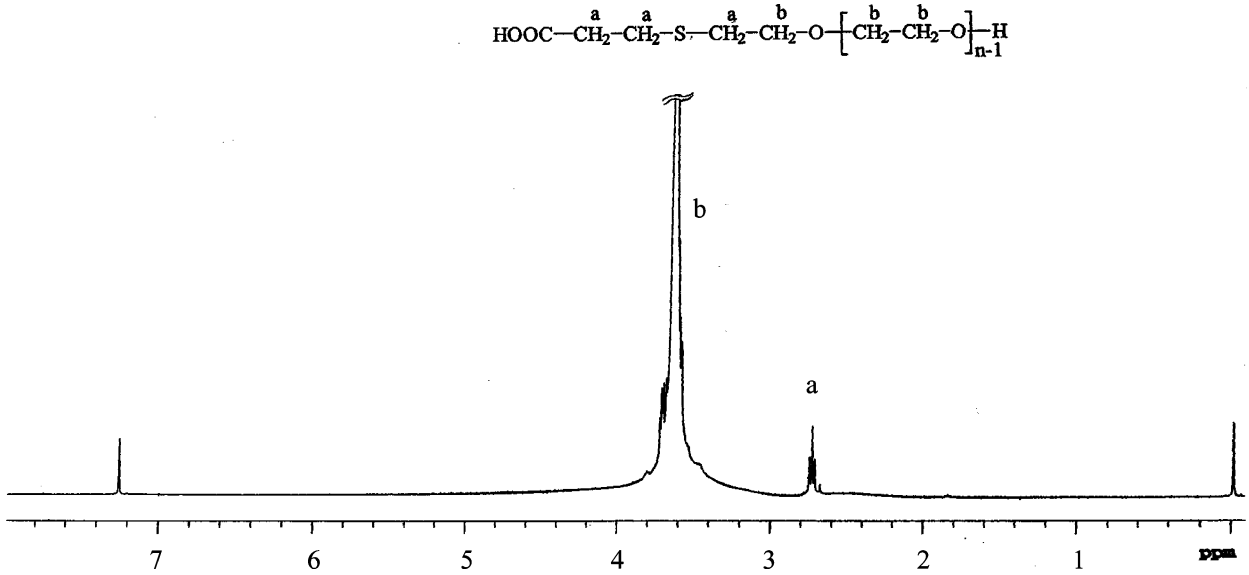


Figure 6. ^{1}H NMR spectrum of carboxy-terminated poly(ethylene glycol): M_n = 2670, M_w/M_n = 1.15 (Table 3, entry 2). M_n = number-average molecular weight; M_w = weight-average molecular weight; M_w/M_n = molecular weight distribution.

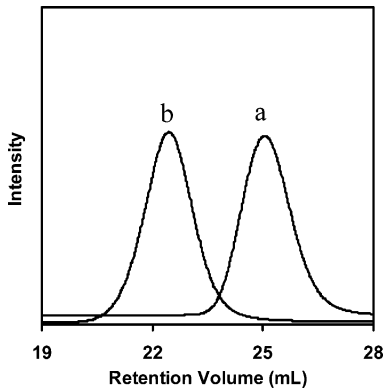


Figure 7. Gel permeation chromatography traces of carboxy-terminated poly(ethylene glycol) and the crude product from the block copolymerization of carboxy-terminated poly(ethylene glycol) with ϵ-caprolactone: (a) $M_n = 2760$, $M_w/M_n = 1.15$ (Table 3, entry 2); (b) $M_n = 11\,000$, $M_w/M_n = 1.16$ (Table 4, entry 2). M_n = number-average molecular weight; M_w = weight-average molecular weight; M_w/M_n = molecular weight distribution.

As shown in Scheme 1, one protected carboxyl and three unprotected carboxyls containing thiol-anionic initiators were evaluated for polymerization of EO.

As summarized in Table 2 (entries 1−3), the *n*-butyl carboxy-protected initiator, K−nBMP, quantitatively initiated EO polymerization to yield polymers with controlled molecular weights ($M_{n,NMR} = 950-5100$) and relatively narrow molecular weight distributions ($M_w/M_n = 1.18-1.25$). In each case the degree of monomer conversion was ≥92% following the 24 h reaction period in THF at 40 °C. Figure 1 includes the GPC trace for the crude reaction product (Table 2, entry 2). The molecular weights of the obtained polymers agreed well with the values calculated from the monomer/initiator ratio. Figure 2 includes the ^{1}H NMR spectrum for one of the polymers prepared by this method (Table 2, entry 2). The resonance at 3.63 ppm was assigned to the protons of the PEG unit (−***CH***$_2$−***CH***$_2$−O and $CH_3-CH_2-CH_2-$***CH***$_2$−O−) and the resonances at 0.91 ppm (***CH***$_3$−$CH_2CH_2-CH_2$−O), 1.38 ppm (CH_3-***CH***$_2$−CH_2-CH_2−O), and 1.55 pm (CH_3-CH_2-***CH***$_2$−CH_2−O) were assigned to the protecting *n*-butyl group. Finally, the resonance at 2.78 ppm was assigned to the methylene group (CO−***CH$_2$CH$_2$***−S-***CH$_2$***) adjacent to the thiol and carbonyl groups. These results indicate the successful incorporation of the protected functional initiator into the resulting PEG polymers. The resonance for the methylene proton adjacent to the carbonyl group and the resonance for the four methylene protons adjacent to the thiol group all appear at 2.78 ppm.

As summarized in Table 2 (runs 4−9), the potassium salts of the unprotected carboxy-functionalized thiol anionic initiators, K−MUDA, K−MPA, and K−MAA, were also used to initiate EO polymerization. The nucleophilicity for these initiators increases in the following order: K−MUDA < K−MPA < K−MAA.

The use of K−MUDA, for EO polymerization in THF at 40 °C, resulted in a PEG with a slightly higher molecular weight than that predicted and GPC analysis revealed a bimodal distribution (Table 2, entries 4 and 5) with a slightly broad molecular weight distribution ($M_w/M_n = 1.2$). However, once MUDA was dried by the azeo-distillation of toluene, a procedure to remove trace amounts of water, the formation of the high molecular weight portion was suppressed (Figure 3A, Table 2, entry 5). This indicates that the high molecular weight fraction of polymer may be produced by reaction of trace amounts of water with potassium naphthalene to yield potassium hydroxide which is also an effective initiator for EO polymerization.[2]

Use of K−MAA for EO polymerization resulted in polymers with a broad molecular weight distribution ($M_w/M_n = 1.20-1.56$, Table 2, entries 6−8) even though the monomer conversion was almost complete within 24 h at 40 °C. GPC analysis indicated that the resulting polymers had a bimodal distribution which may be attributed to the high nucleophilicity of this initiator. In contrast, when K−MPA was used as the initiator, the molecular weight of the resulting polymer was as predicted from the monomer/initiator ratio and the molecular weight distribution was narrow (i.e., $M_w/M_n = 1.07$, Table 2, entry 9). The GPC trace for the polymer synthesized by this method is shown in Figure 3B. Since these preliminary results identified K−MPA as an efficient initiator for EO polymerization (i.e., initiator efficiency > 90%), it was used to prepare a series of PEG polymers as summarized in Table 3.

For all functionalized PEGs synthesized using K−MPA the monomer conversion was higher than 80%, the molecular weights of the polymers agreed well with values predicted from the monomer/initiator ratios ($M_{n,cal}$) and the molecular weight distributions of the polymers were narrow ($M_w/M_n \leq 1.15$). The initiator efficiencies were as high as 0.92, which is higher than the initiator efficiencies for most of the carboxy-functionalized oxy-anionic initiators (Table 1). Most importantly, this method

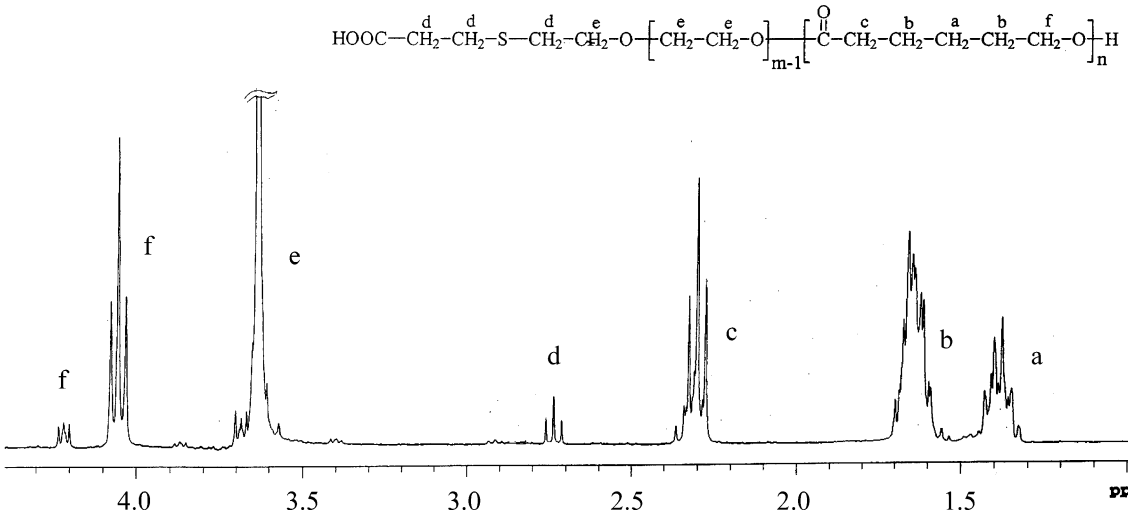


Figure 8. ^{1}H NMR spectrum of carboxy-terminated poly(ethylene glycol)-*block*-poly(ϵ-caprolactone) ($M_n = 11\,000$, $M_w/M_n = 1.15$, Table 4, entry 2). M_n = number-average molecular weight; M_w = weight-average molecular weight; M_w/M_n = molecular weight distribution.

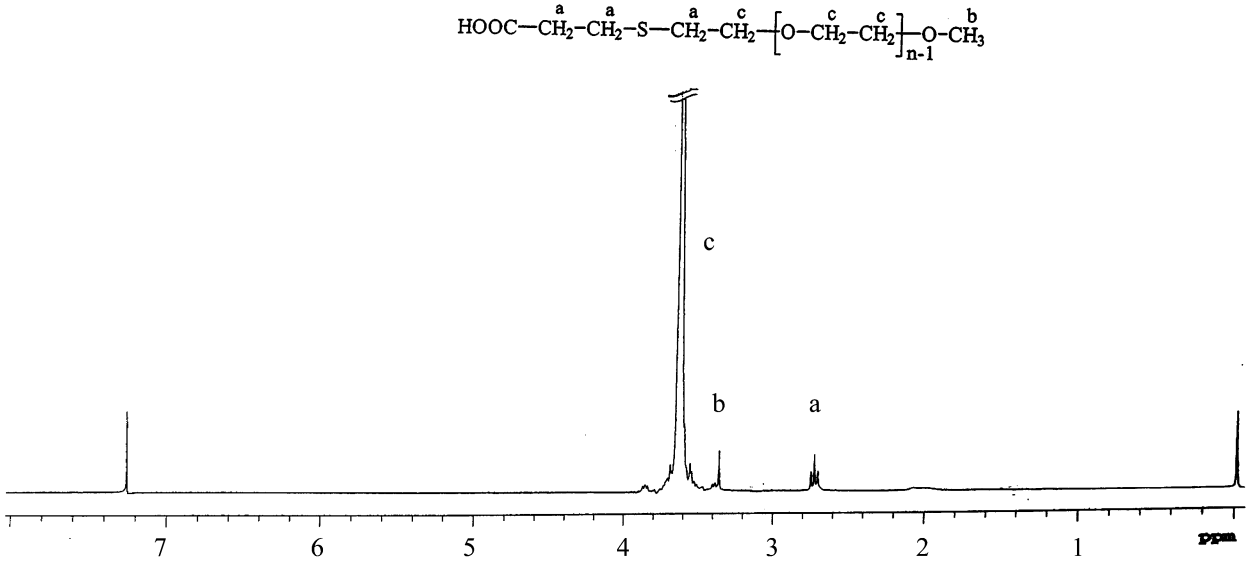


Figure 9. ^{1}H NMR spectrum of α-carboxy-ω-methoxy−poly(ethylene glycol) (HOOC−PEG−OCH_3, Table 3, entry 3a, M_n = 7190, M_w/M_n = 1.09). M_n = number-average molecular weight; M_w = weight-average molecular weight; M_w/M_n = molecular weight distribution.

allowed for preparation of PEG with molecular weights as high as $M_{n,GPC}$ = 24 700 (Table 3, entry 6) and a narrow molecular weight distribution ($M_w/M_n \leq 1.15$), indicating that this thiol anionic initiator can be used for preparation of high molecular weight PEG. As shown in Figure 4, the molecular weights of the polymer increase linearly while the molecular weight distributions of the polymer slightly decrease with increasing monomer/initiator ratio. Figure 5 includes the GPC traces for PEGs prepared from increasing ratios of monomer to initiator (i.e., $[EO]_0/[MPA]_0$). In an attempt to prepare PEG with a molecular weight of 20 000 the resulting product (trace e) includes a high molecular weight fraction which amounted to approximately 12% of the total product, as determined by GPC. This high molecular weight fraction likely results from the presence of trace amounts of water in the reaction mixture.

The syntheses of the carboxy-terminated heterobifunctional PEGs were confirmed by ^{1}H NMR analyses. Figure 6 includes a sample ^{1}H NMR spectrum for a HOOC−PEG−OH polymer (Table 3, entry 2). The resonance corresponding to the PEG unit are at 3.63 ppm (O−***CH***$_2$−***CH***$_2$−) and the resonance at 2.74 ppm is assigned to the methylene proton adjacent to the thiol and carboxyl groups (6H, −***CH***$_2$−S−***CH***$_2$−**C**H_2−COOH), which is similar to the corresponding protons from nBMP-terminated PEG.

3. Synthesis of Block Copolymers of Carboxy-Functionalized Poly(ethylene glycol)-*b*-poly(ϵ-caprolactone). To demonstrate the utility of the HOOC−PEGs for preparation of copolymers they were employed as macroinitiators (Table 3, entries 2−3) for polymerization of CL using the recently developed acid-based (HCl-ether catalyst) cationic polymerization method.[24] A series of PEG-*b*-PCL copolymers were successfully synthesized as summarized in Table 4. The monomer conversion of CL was almost complete within a 24 h period and the composition of the copolymers, as determined from ^{1}H NMR analysis, were as predicted. GPC analysis of the copolymers revealed relatively narrow molecular weight distributions (M_w/M_n < 1.16). Figure 7 includes a typical GPC trace for a HOOC−PEG-*b*-PCL from the crude reaction mixture (Table 4, entry 1). As shown, according to GPC analysis, no residual amount of unreacted PEG macroinitiator remained following polymerization which also highlights the high functionality of the macroinitiators. Figure 8 includes a typical ^{1}H NMR spectrum for the HOOC−PEG-*b*-PCL copolymer (Table 4, entry 2). The resonance at 3.63 ppm was assigned to the PEG unit (***CH***$_2$−**CH**$_2$−O) and resonance for the PCL unit appeared at 1.35 ppm (m, 2H, CO−CH_2−CH_2−***CH***$_2$−CH_2−CH_2−O), 1.55 ppm (m, 4H, CO−CH_2−***CH***$_2$−CH_2−***CH***$_2$−CH_2−O), 2.28 ppm (m, 2H, CO-***CH***$_2$−CH_2−CH_2−CH_2−CH_2−O), 4.07 ppm (m, 2H, CO−CH_2−CH_2−CH_2−CH_2−***CH***$_2$−O). More importantly, the resonance at 2.76 ppm is assigned to the methylene proton adjacent to the thiol and carboxyl groups (6H, −***CH***$_2$−S-***CH***$_2$−**CH**$_2$−COOH), which indicates the successful synthesis of the HOOC−PEG-*b*-PCL.

4. Synthesis of α-Carboxy-ω-Methoxy PEG. Scheme 2 also includes the procedure for synthesis of α-carboxy-ω-methoxy PEG wherein MeI was used to terminate the living polymer chain of PEG. Following the addition of MeI, the reaction mixture gradually became heterogeneous, indicating the production of potassium iodide. The results for synthesis with termination using MeI are summarized in Table 3 and a typical ^{1}H NMR spectrum, with assignments as shown, is included in Figure 9 (Table 3, entry 3a). Overall, the results demonstrate that this is an efficient method for synthesizing the heterobifunctional PEG containing a reactive carboxyl group at one terminus and the nonreactive methoxy group at the other terminus.

Summary

Potassium-3-mercaptopropionic acid is an effective functional initiator for synthesis of heterobifunctional PEGs. Use of this initiator allows for controlled polymerization of EO up to molecular weights of 20 000 and yields polymers with narrow molecular weight distributions. The α-carboxy-ω-hydroxy PEG was successfully used as the macroinitiator for synthesis of a series of α-carboxy−PEG-*b*-PCL copolymers by an HCl catalyzed cationic polymerization method. The α-carboxy-ω-methoxy−PEG was also synthesized using MeI as the termination agent.

Acknowledgment. The authors thank the National Science and Engineering Research Council (NSERC) for supporting this research.

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MA0607665