Well-Defined Oligosaccharide-Terminated Polymers from Living Radical Polymerization

David M. Haddleton* and Kohji Ohno

Department of Chemistry, University of Warwick, Coventry CV4 7AL, U.K.

Received March 17, 2000; Revised Manuscript Received April 6, 2000

β-cyclodextrin has been transformed into an initiator for copper(I)-mediated living radical polymerization via successive acetylation, ring opening, and condensation reactions. This glycoinitiator has been successfully used to prepare a range of methacrylate polymers, which show all characteristics of living polymerization. Poly(methyl methacrylate) is prepared with an M_n of 10 100 g/mol (theoretical M_n = 10 900) and polydispersity of 1.09. Excellent first-order kinetics and an evolution of mass with time are presented. Hydrophilic polymers based on poly(ethylene glycol), glucose and tertiary amine monomers are successfully prepared with terminal maltoheptose units. Polymerization of styrene results in a broadening of the polydispersity to 1.48 while maintaining good control over the M_n. The acetyl protecting groups on the maltoheptose terminal unit are easily removed in all cases to give the hydroxylated sugar, as shown by NMR. These results demonstrate the applicability of utilizing glycoinitiators as a new, versatile route to a wide range of glycopolymers. The polymerization chemistry is inert to the sugar functionality allowing glyco units to be precisely placed within a synthetic macromolecule with all the associated advantages of living polymerization.

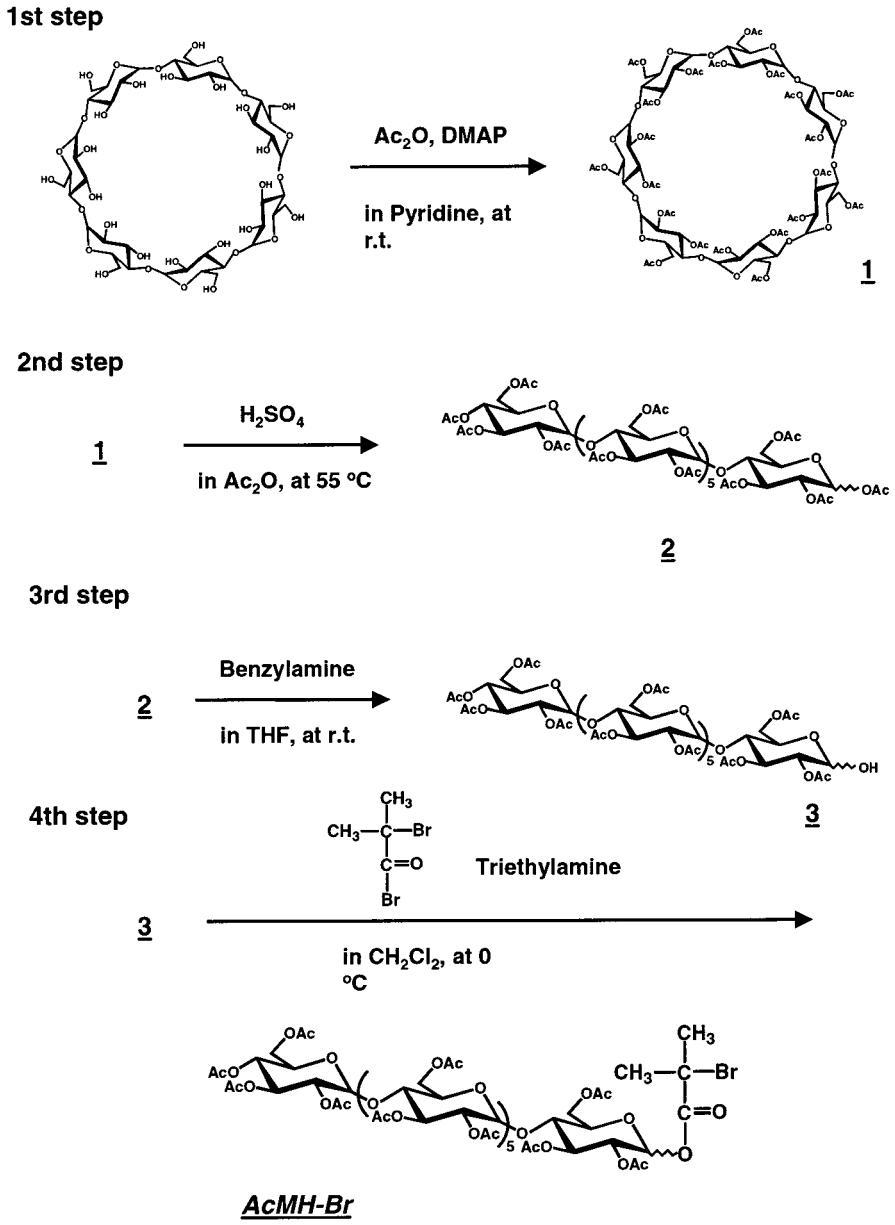
Introduction

There is an increasing interest in polymers having sugar residues, *glycopolymers*, as a result of their potential advantageous physicochemical and structural properties.[1,2] Naturally occurring polysaccharides are present in a range of glycoproteins, glycolipids, etc. and play key roles in a wide range of biochemistry as a result of excellent biorecognition properties. Synthetic polymers that contain sugar moieties can be expected to act as model compounds with a wide range of differing properties depending upon the comonomers utilized. To date, many synthetic glycopolymers have been synthesized by conventional free radical polymerizations of vinyl monomers with pendant saccharide residues.[1] Conventional free radical polymerization of a glycomonomer with appropriate comonomers yields polymers where the sugar functionality is statistically distributed along the chain according to the reactivity ratios of the monomer pair. Such comonomers can range from being extremely hydrophilic to hydrophobic in nature and from being functional or nonfunctional with all intermediary values easily attainable. Even where the biorecognition site is statistically distributed useful biofunctionality has been demonstrated. Polymers containing maltoheptose units as side chains from poly(methyl methacrylate) have been demonstrated to have anti-HIV activity and blood anticoagulant activity.[3a] Other synthetic vinyl polymers containing statistical distributions of D-glucaric residues have been shown to inhibit the activity of β-D-glucuronidase.[3b] Thus, the construction of synthetic glycoconjugates which can potentially participate in a range of processes by the attachment of sugars to polymeric molecules is of significant current interest. Recently well-defined, low-polydispersity glycopolymers have been achieved by several novel polymerization methods[4] including controlled/"living" radical polymerizations.[5] The use of controlled and living radical polymerization methods allows for the sugar containing part of the molecule to be precisely placed within the macromolecule as blocks, termini, etc.[6] This offers an attractive advantage over traditional methods in that the exact location of a recognition site within a macromolecule has important consequences with regards bioactivity. The use of living radical and in particular transition-metal-mediated living radical polymerization offers an extremely robust synthetic protocol which is inert to many different types of initiators and various types of monomers.[7] In our laboratory, we have been developing catalysts for living radical polymerization based on copper(I) halides and alkylpyridylmethanimine ligands.[8] We have also been developing the synthesis of functional initiators and monomers so as to prepare well-defined polymers with novel effects and properties.[9] Our current interest has focused on the synthesis of carbohydrate-based polymers by living radical polymerization in which carbohydrates have been used to synthesize initiators and monomers in order to prepare synthetic polymers with well-defined sugar functionality at specific places within each macromolecule. Herein we report the use of an oligosaccharide-carrying initiator suitable for living radical polymerization as derived from β-cyclodextrin (β-CD) in order to precisely synthesize a novel type of glycopolymer, viz. a block copolymer consisting of a maltoheptose terminus and a well-defined, low-polydispersity synthetic poly(methacrylate). The focus of the current paper is to report a versatile approach to glycopolymers, which has the potential to be used for all vinyl-containing gly-

* Corresponding author. Fax: (+44)2476-524112. E-mail: D.M.Haddleton@warwick.ac.uk.

10.1021/bm005531u CCC: $19.00
Published on Web 05/05/2000

Scheme 1. Synthesis Route to 1-*O*-(2-Bromo-2-methylpropionyl) Maltoheptaoside, AcMH−Br



comonomers and sugar-based initiators as derived from condensation of hydroxyl functionality with appropriate acyl halides as described herein. As such the chemistry has the potential to be widely applied. The example used is based on β-cyclodextrin, a readily available and inexpensive sugar residue.

Results and Discussion

A four-step reaction (Scheme 1) was carried out from commercially available β-CD to prepare the oligosaccharide with protected hydroxyl groups carrying an initiator for living radical polymerization, AcMH−Br. The final product was purified by chromatography (silica gel) so as to obtain an analytically pure compound. Living radical polymerization of methyl methacrylate (MMA) was carried out in the presence of an appropriate copper(I) complex AcMH−Br as initiator. Figure 1 shows size exclusion chromatography (SEC) curves of the initiator AcMH−Br and the PMMA thus obtained as a function of time and conversion. The molecular mass of the PMMA increases with increasing reaction time with no detectable shoulder peak at the elution position of the initiator after 15 min reaction time, the M_w/M_n ratio remains remarkably low in all cases, where M_w and M_n are the weight- and number-average molecular masses, respectively. Figure 2 shows a first-order plot for the reaction. The plot is linear, passing through the origin, indicating that the concentration of propagating macromolecules stays constant throughout the polymerization, interestingly about 90% conversion is reached after 2 h. Figure 3 shows the evolution of M_n and M_w/M_n of the PMMA as a function of monomer conversion. The M_n increases fairly linearly with conversion remaining close to the theoretical value, $M_{n,theo}$, ($M_{n,theo} = M_{MMA}([MMA]/[AcMH-Br]) \times$ % conversion). This indicates that the initiator efficiency of AcMH−Br is approximately 100%. The Polydispersity, M_w/M_n, stays below 1.2 throughout the reaction. These results confirm that the

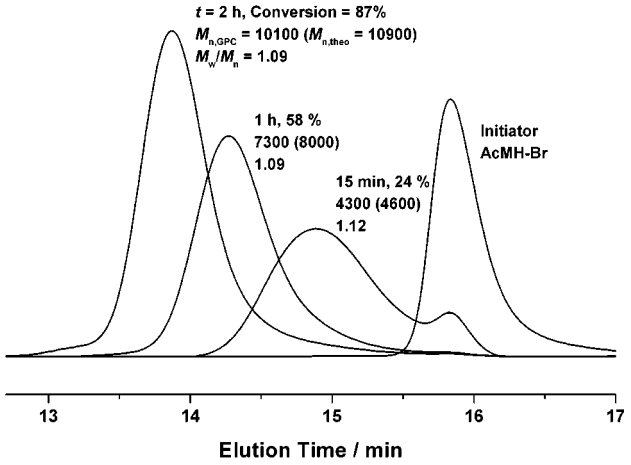


Figure 1. SEC curves for the polymerization of MMA in toluene at 90 °C: $[MMA]_0/[AcMH-Br]_0/[Cu(I)Br]_0 = 100/1/1$.

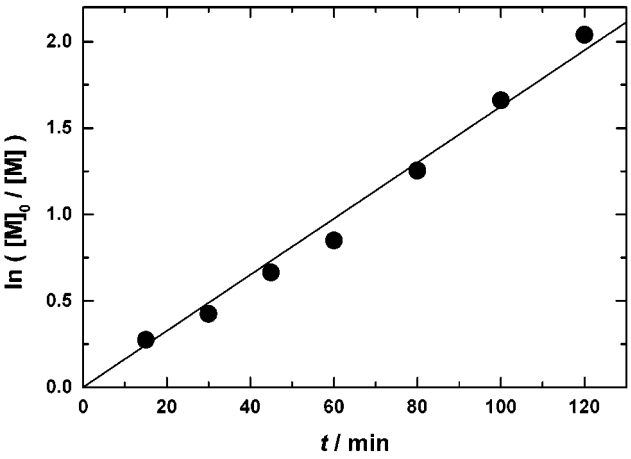


Figure 2. First-order plot for the solution polymerization of MMA in toluene at 90 °C: $[MMA]_0/[AcMH-Br]_0/[Cu(I)Br]_0 = 100/1/1$.

polymerization proceeds in a "living" fashion with negligible contribution of termination and transfer reactions.

The resulting polymers were treated with sodium methoxide in a mixture of chloroform and methanol to deprotect the acetyl groups from the initiator moiety so as to obtain PMMA having an oligosaccharide residue at the α-chain terminus. Figure 4 shows typical ^{1}H NMR spectra of the oligosaccharide-terminated PMMA taken before (a) and after (b) the hydrolysis. The signals of acetyl group protons (1.9−2.2 ppm in Figure 4a) have completely disappeared after the hydrolysis. Furthermore, the signals correspondent to polymer backbone and sugar residue can be observed in the spectrum even after hydrolysis. These results confirm that the deprotection of the acetyl groups proceeds quantitatively while leaving the methyl ester groups of the polymer backbone and the initiator unit unaffected.

These excellent results encouraged us to apply this system to the polymerizations of various functional methacrylates; see Table 1. Co-methacrylate monomers were chosen so as to produce water-soluble polymers containing both cationic and neutral hydrophilic functionality. The range of functionality within the monomers serves to demonstrate the applicability of this approach to methacrylates in general. All methacrylate derivatives used here, including hydrophilic (DMAEMA), hydrophilic macromonomer (PEGMA), and sugar-carrying monomer (MAIpGlc), Scheme 2, were

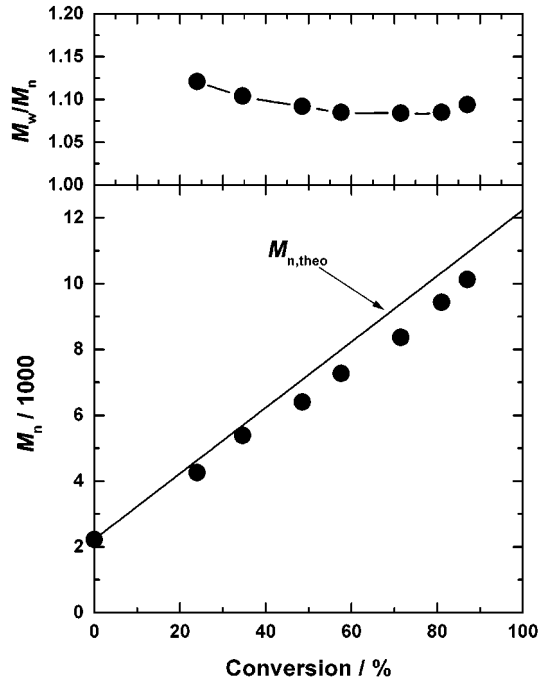


Figure 3. Evolution of M_n and M_w/M_n as a function of monomer conversion for the solution polymerization of MMA in toluene at 90 °C: $[MMA]_0/[AcMH-Br]_0/[Cu(I)Br]_0 = 100/1/1$. The full line in the figure represents the theoretical prediction.

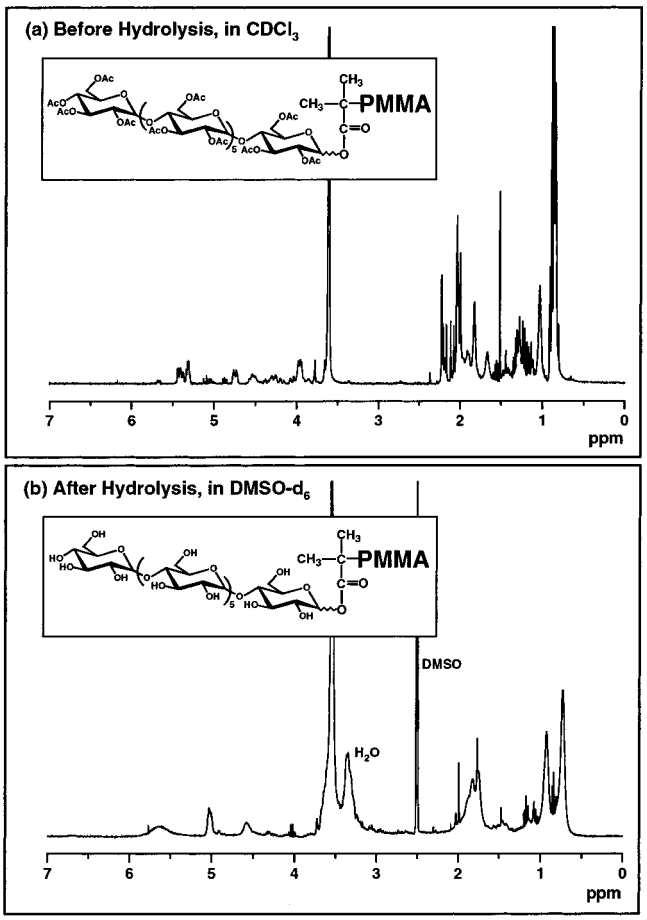


Figure 4. ^{1}H NMR spectra taken (a) before and (b) after the hydrolysis of AcMH−PMMA. (a) $CDCl_3$ and (b) DMSO-d_6.

polymerized with good control, giving low polydispersity polymers. Unfortunately, reliable information about

Table 1. Polymerization Data for the Living Radical Polymerization Initiated with AcMH−Br[a]

monomer	$[M]_0/[I]_0/[Cu]_0/[L]_0$[b]	T/°C	t/h	convn/%	$M_{n,GPC}$	$M_{n,theor}$[e]	M_w/M_n
MMA	100/1/1/2	90	2	87	10 100[c]	10 900	1.09[c]
ST	70/1/1/2	110	15	91	10 700[d]	8 900	1.48[d]
PEGMA	30/1/1/2	90	72	80	11 500[c]	13 600	1.15[c]
MAIpGlc	80/1/1/2	90	21	88	16 500[c]	25 300	1.21[c]
DMAEMA	70/1/1/2	90	8	82		11 300	

[a] Concentration of monomer = 50 wt %. Toluene was used as the solvent except for styrene, for which *p*-xylene was used. [b] $[M]_0$, $[I]_0$, $[Cu]_0$, and $[L]_0$ represent initial concentrations of monomer, AcMH−Br, Cu^IBr, and ligand, respectively. [c] Estimated by PMMA-calibrated SEC. [d] Estimated by PST-calibrated SEC. [e] $M_{n,theo} = M_{MMA}([MMA]/[AcMH-Br]) \times$ % conversion.

PDMAEMA could not be obtained by our current SEC system, presumably due to some interaction between the polymer and the column resin. The backbone isopropylidene-protected glycopolymer (PMAIpGlc) was treated with formic acid to obtain a novel type of well-defined, water-soluble glycopolymer with sugar residues both along the backbone and at the chain end. In the case of styrene (ST) polymerization, the polydispersity of the product broadens, but the molecular mass remains close to the theoretical value. This is similar to that observed when living radical polymerization of styrene is carried out using other types of 2-bromo-2-methylpropionyl initiator.[9]

Summary

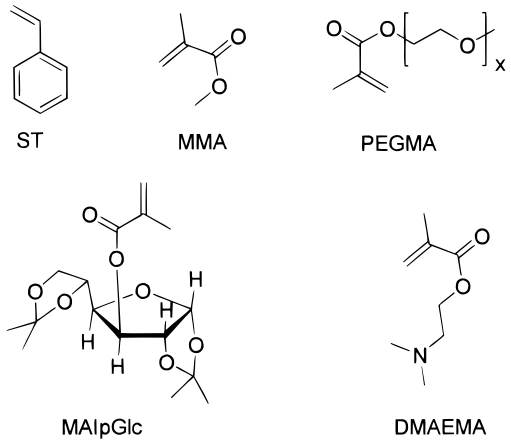
In conclusion, a glyco-derived initiator with a peracetylated oligosaccharide residue is an effective initiator for copper(I)-mediated living radical polymerization of methacrylates to give low-polydispersity polymers with expected molecular masses. The system is applicable to various methacrylate derivatives including hydrophilic monomer and macromonomer, resulting in good control of the polymerization. The hydrolysis of the obtained polymers provided various types of oligosaccharide-terminated polymers with well-defined structures. Thus, the present work provides a route to well-defined polymers with an oligosaccharide residue. The obtained polymers would be expected to express particular solution or solid properties that would arise from amphiphilic character, and some biological function due to the particular properties of sugar residues. Studies on these properties are underway now and will be the topic of a forthcoming full paper.

Experimental Section

Reagents: β-Cyclodextrin hydrate (Avocado) was dried in vacuo at 80 °C with phosphorus pentoxide overnight immediately prior to use. Styrene, methyl methacrylate, and dimethylaminoethyl methacrylate (DMAEMA) were obtained from Aldrich and purified by passing through a column of activated basic alumina to remove inhibitor. Sugar-carrying monomer, MAIpGlc, was synthesized according to the method described previously.[5b] Poly(ethylene glycol) methyl ether methacrylate (PEGMA, average M_n ca. 475) was used as received from Aldrich. All other reagents were commercially obtained and used without further purification.

Measurements. The SEC analysis was carried out on a system equipped with a guard column and two 30 cm mixed C column (Polymer Laboratories) with differential refractive index detection using tetrahydrofuran (THF) at 1 mL/min as an eluent. Poly(methyl methacrylate) (PMMA) and poly(styrene) (PST) standards were used to calibrate the SEC system. ^{1}H NMR and ^{13}C NMR spectra were recorded on a Bruker AC300 and a Bruker AC400 spectrometer.

Scheme 2. Monomers Used in This Work



Synthesis of 1-*O*-(2-Bromo-2-methylpropionyl) Maltoheptaoside (AcMH−Br, Scheme 1). AcMH−Br was synthesized via a four-step reaction. (1) β-cyclodextrin (β-CD) was acetylated with acetic anhydride in pyridine in the presence of catalytic amount of 4,4′-(dimethylamino)pyridine, giving peracetylated β-CD, **1**, in quantitative yield. (2) **1** was treated with a mixture of acetic anhydride/concentrated sulfuric acid (49:1, *v*:*v*) at 55 °C for 20 h, giving *O*-acetylmaltoheptaose, **2**, in 40−50% yield.[10] (3) **2** was treated with benzylamine in THF for 24 h at room temperature to selectively deacetylate at the reducing end of the oligosaccharide, providing peracetylated maltoheptaose having a hydroxyl group at the reducing end, **3**, in 60−70% yield.[3a] (4) To a cold solution of **3** (4 g, 1.94 mmol) in dry dichloromethane (20 mL) with triethylamine (0.4 mL, 2.91 mmol) was added 2-bromoisobutyryl bromide (0.48 mL, 2.91 mmol) dropwise at 0 °C. The mixture was magnetically stirred for 2 h at 0 °C and then for 4 h at room temperature. The system was diluted with dichloromethane (170 mL) and washed successively with 1 N aqueous HCl solution (200 mL), 5% aqueous $NaHCO_3$ (200 mL), and pure water (200 mL × 2). The organic layer was dried over anhydrous sodium sulfate. After the solvent was removed, the residue was purified by flash silica gel column chromatography with a 2:5 toluene/ethyl acetate mixture as an eluent and then was recrystallized from ethanol to yield a white powder as a final product (3.15 g, 73%). ^{13}C NMR ($CDCl_3$, 75 MHz): δ 20.6, 20.7, and 20.9 (CH_3CO), 30.1 and 30.3 (α-CH_3), 54.6 (*C*Br), 60.4−62.6, 67.9−74.8, 92.5, and 95.7 (sugar carbons),

169.5−169.9 (C═O). Anal. Calcd for $C_{90}H_{121}O_{59}Br$: C, 48.54; H, 5.49. Found: C, 47.61; H, 5.41.

Typical Polymerization Procedure. A Schlenk flask was charged with a predetermined amount of Cu(I)Br, to which a mixture of monomer, solvent (toluene or *p*-xylene), AcMH−Br, and *n*-propyl-2-pyridylmethanimine, as a bidentate ligand for copper complexation, was quickly added. The mixture was immediately degassed by three freeze−pump−thaw cycles and then purged under a nitrogen atmosphere. The polymerization reaction was carried out in an oil bath thermostated at a predetermined temperature for a prescribed time while stirring. As for kinetic experiments, the polymerization solution was sampled at suitable time period throughout the reaction by a gastight syringe. The sample solution was passed through a short alumina column to remove the copper complexes. Molecular mass and molecular weight distribution were determined by SEC. Monomer conversion was estimated by gravimetry by taking to constant weight in a vacuum oven at 50 °C. Final polymer products were further purified by reprecipitation with a dichloromethane/petroleum ether system and dried in a vacuum oven.

Deprotection of Polymers with Acetylated-Oligosaccharide Residue: A solution of PMMA−AcMH (620 mg) in $CHCl_3$ (10 mL), MeOH (10 mL) and 1 M $NaOCH_3$ in MeOH (150 mg) was stirred for 1 h at room temperature. After neutralization of the system with a small amount of Amberlite IR-120 (H^+ form) and then filtration, the solvent was evacuated off. The resulting polymer was further dried in vacuo.

Acknowledgment. We would like to thank EPSRC for funding (K.O.; GR/M45634).

References and Notes

(1) For recent review, see: (a) Wulff, G.; Schmid, J.; Venhoff, T. *Macromol. Chem. Phys.* **1996**, *197*, 259. (b) Miyata, T.; Nakamae, K. *Trends Polym. Sci.* **1997**, *5*, 198.

(2) (a) Kallin, E.; Lönn, H.; Norberg, T.; Elofsson, M. *J. Carbohydr. Chem.* **1989**, *8*, 597. (b) Kobayashi, K.; Kobayashi, A.; Tobe, S.; Akaike, T. In *Neoglycoconjugate: Preparation and Application*; Lee, Y. C., Lee R. T., Eds.; Academic Press: San Diego, CA, 1994; p 261. (c) Kobayashi, K.; Akaike, T.; Usui, T. In *Methods in Enzymology*; Lee, Y. C., Lee R. T., Eds.; Academic Press: San Diego, CA, 1994; Vol. 242, p 226. (d) Nishimura, S.-I.; Matsuoka, K.; Furuike, T.; Nishi, N.; Tokura, S.; Nagami, S.; Murayama, S.; Kurita, K. *Macromolecules* **1994**, *27*, 157. (e) Roy, R.; Tropper, F. D.; Romanowska, A. *Bioconjugate Chem.* **1992**, *3*, 256. (f) Roy, R.; Andersson, F. O.; Harms, G.; Kelm, S.; Schauer, R. *Angew. Chem., Int. Ed. Engl.* **1992**, *31*, 1478. (g) Spaltenstein, A.; Whitesides, G. M. *J. Am. Chem. Soc.* **1991**, *113*, 686.

(3) (a) Yoshida, T.; Akasaka, T.; Choi, Y.; Hattori, K.; Yu, B.; Mimura, T.; Kaneko, Y.; Nakashima, H.; Aragaki, E.; Premanathan, M.; Yamamoto, N.; Uryu, T. *J. Polym. Sci., Part A: Polym. Chem.* **1999**, *37*, 789. (b) Hashimoto, K.; Ohsawa, R.; Imai, N.; Okada, M. *J. Polym. Sci., Part A: Polym. Chem.* **1999**, *37*, 303.

(4) (a) Yamada, K.; Yamaoka, K.; Minoda, M.; Miyamoto, T. *J. Polym. Sci., Part A: Polym. Chem.* **1997**, *35*, 255. (b) Yamada, K.; Minoda, M.; Miyamoto, T. *J. Polym. Sci., Part A: Polym. Chem.* **1997**, *35*, 751. (c) Fraser, C.; Grubbs, R. H. *Macromolecules* **1995**, *28*, 7248. (d) Aoi, K.; Suzuki, H.; Okada, M. *Macromolecules* **1992**, *25*, 7073. (e) Aoi, K.; Tsutsumiuchi, K.; Okada, M. *Macromolecules* **1994**, *27*, 875. (f) Loykulnant, S.; Hayashi, M.; Hirao, A. *Macromolecules* **1998**, *31*, 9121.

(5) (a) Ohno, K.; Tsujii, Y.; Miyamoto, T.; Fukuda, T.; Goto, M.; Kobayashi, K.; Akaike, T. *Macromolecules* **1998**, *31*, 1064. (b) Ohno, K.; Tsujii, Y.; Fukuda, T. *J. Polym. Sci., Part A: Polym. Chem.* **1998**, *36*, 2473. (c) Ohno, K.; Fukuda, T.; Kitano, H. *Macromol. Chem. Phys.* **1998**, *199*, 2193. (d) Ohno, K.; Izu, Y.; Yamamoto, S.; Miyamoto, T.; Fukuda, T. *Macromol. Chem. Phys.* **1999**, *200*, 1619.

(6) Matyjaszewski, K. *Chem.−Eur. J.* **1999**, *5*, 3095.

(7) (a) Kato, M.; Kamigaito, M.; Sawamoto, M.; Higashimura, T. *Macromolecules* **1995**, *28*, 1721. (b) Wang, J. S.; Matyjaszewski, K. *J. Am. Chem. Soc.* **1995**, *117*, 5614. (c) Percec, V.; Barboiu, B. *Macromolecules* **1995**, *28*, 7970. (d) Hawker, C. J. *Acc. Chem. Res.* **1997**, *30*, 373. (e) Fischer, H. *Macromolecules* **1997**, *30*, 5666. (f) Ashford, E. J.; Naldi, V.; O'Dell, R.; Billingham, N. C.; Armes, S. P. *Chem. Commun.* **1999**, 1285. (g) Zhang, X.; Matyjaszewski, K. *Macromolecules* **1999**, *32*, 7349. (h) Miller, P. J.; Matyjaszewski, K. *Macromolecules* **1999**, *32*, 8760. (i) Pyun, J.; Matyjaszewski, K. *Macromolecules* **2000**, *33*, 217.

(8) (a) Haddleton, D. M.; Jasieczek, C. B.; Hannon, M. J.; Shooter, A. J. *Macromolecules* **1997**, *30*, 2190. (b) Haddleton, D. M.; Waterson, C.; Derrick, P. J.; Jasieczek, C.; Shooter, A. J. *Chem. Commun.* **1997**, 683. (c) Haddleton, D. M.; Heming, A. M.; Kukulj, D.; Duncalf, D. J.; Shooter, A. J. *Macromolecules* **1998**, *31*, 2016. (d) Haddleton, D. M.; Clark, A. J.; Duncalf, D. J.; Heming, A. H.; Kukulj, D.; Shooter, A. J. *J. Mater. Chem.* **1998**, 1525. (e) Haddleton, D. M.; Kukulj, D.; Duncalf, D. J.; Heming, A. H.; Shooter, A. J. *Macromolecules* **1998**, *31*, 5201.

(9) (a) Haddleton, D. M.; Edmonds, R.; Heming, A. M.; Kelly, E. J.; Kukulj, D. *New J. Chem.* **1999**, *23*, 477. (b) Haddleton, D. M.; Waterson, C. *Macromolecules* **1999**, *32*, 8732. (c) Jankova, K.; Chen, X.; Kops, J.; Batsberg, W. *Macromolecules* **1998**, *31*, 538. (d) Angot, S.; Murthy, K. S.; Taton, D.; Gnanou, Y. *Macromolecules* **1998**, *31*, 6748. (e) Leduc, M. R.; Hayes, W.; Fréchet, J. M. J. *J. Polym. Sci., Part A: Polym. Chem.* **1998**, *36*, 1. (f) Patten, T. E.; Matyjaszewski, K. *Acc. Chem. Res.* **1999**, *32*, 895.

(10) Sakairi, N.; Wang, L.; Kuzuhara, H. *J. Chem. Soc., Chem. Commun.* **1991**, 289.

BM005531U