Synthesis of Peptide-Based Polymers by Microwave-Assisted Cycloaddition Backbone Polymerization

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Received October 20, 2006

The optimized reaction conditions for the Cu(I)-catalyzed *N*→*C* polymerization of azido-phenylalanyl-alanyl-propargyl amide to yield either high molecular weight linear polymers or medium-sized cyclic polymers is described. These reaction conditions will be applied to tailor the synthesis, properties, and structure of biologically relevant peptide-based biopolymers.

Introduction

There is great interest in the use of functional peptides as building blocks for the synthesis of peptide-based polymers, since these polymers can be applied for a variety of purposes such as drug delivery systems, scaffolds for tissue engineering and repair, and as novel biomaterials.[1] Nowadays, the majority of such peptide-based polymers are hybrids of *N*-terminally grafted peptides to a synthetic polymer backbone.[2] However, peptide-based biopolymers derive their structural, mechanical, and biological properties from their backbone sequential repetitions rather than by grafting the peptide sequence to the polymeric backbone. Examples of peptide biopolymers containing repetitive sequences are, among others, collagens,[3] elastin and spider silk,[4] antifreeze proteins,[5] mussel glue,[6] and reflectins.[7]

Current methods for the synthesis of amino acid-based polymers use amino acid-based *N*-carboxy anhydrides[8,9] for the synthesis of poly-Glu or poly-Lys, activated peptide esters toward the synthesis of polymeric elastin models[10] or condensing agents such as diphenylphosphorylazide,[5,11] carbodiimides, and acid chlorides[12] (see, for a review, ref 2b).

The reaction between terminal acetylenes and organic azides yielding the corresponding 1,4-disubstituted 1,2,3-triazoles[13] catalyzed by copper (I) seems particularly suitable for chemoselective conjugation reactions. So far, this 1,3-dipolar cycloaddition reaction denoted as the "click reaction"[14] has led to a plethora of applications in the literature.[14,15] Recent studies showed that the 1,2,3-triazole moiety is an effective mimic of a peptide amide bond as present in a β-strand[15d] or as a dipeptide replacement in α-helical coiled coils.[16] The possibility of chemoselective azide−alkyne coupling in the presence of other (unprotected) functional groups and these topological similarities between peptide amides and 1,2,3-triazoles made us decide to explore the 1,3-dipolar cycloaddition reaction for the synthesis of peptide-based polymers.[17]

Here we describe that the model dipeptide azido-phenylalanyl-alanyl-propargyl amide **1** (Figure 1) can be efficiently converted into high molecular weight amino acid-based polymers (up to 45 000 Da) by a microwave-assisted 1,3-dipolar cycloaddition reaction. Depending on the reaction conditions, we found that the outcome of the click reaction can be directed either to large linear polymers (up to 300 amino acid residues) or medium-sized peptide macrocycles (4−20 amino acid residues).

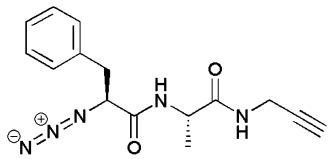

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Figure 1. Structure of azido-phenylalanyl-alanyl-propargyl amide.

Experimental Section

Monomer Synthesis. Full experimental details of the monomer synthesis and the analysis of the corresponding intermediates are given in the Supporting Information section. Compound **1** was obtained as a white solid in 71% yield (2.13 g, 7.1 mmol). $R_f = 0.61$ ($CHCl_3$/MeOH/AcOH 95:20:3 v/v/v); mp: 91 °C; Fourier transform infrared (FTIR) (KBr) υ: 2100 cm^{-1}; ^1H NMR ($CDCl_3$, 300 MHz) δ: 1.27 (d, 3H, βCH_3 Ala), 2.23 (s, 1H, C≡CH), 3.06−3.30 (dd (J_{ax} 13.9 Hz, J_{bx} 121 Hz), 2H, βCH_2 Phe), 4.02 (m, 2H, CH_2, $CH_2C{\equiv}CH$), 4.19 (m, 1H, αCH Ala), 4.48 (m, 1H, αCH Phe), 6.78 (broad t, 1H, NH), 6.88 (d, 1H, NH Ala), 7.22−7.35 (m, 5H, arom H). ^{13}C NMR ($CDCl_3$, 75 MHz) δ: 11.2 (βCH_3 Ala), 22.5 ($CH_2C{\equiv}CH$), 31.6 (βCH_2 Phe), 41.8 (αCH Ala), 58.2 (αCH Phe), 65.0 (C≡*C*H), 72.3 (*C*≡CH), 120.6, 121.9, 122.6, 128.9 (arom C), 162.0 (CO Ala), 164.5 (CO Phe); Anal. Calcd for $C_{15}H_{17}N_5O_2$: C 60.12%, H 5.72%, N 23.40%; found: C 60.04%, H 5.65%, N 23.24%.

Polymer Synthesis. A complete list of all polymerization conditions and the analysis of the isolated polymers is given in the Supporting Information section, Table S1. In a typical experiment (entry 5), monomer **1** (1000 mg, 3.34 mmol) was dissolved in N_2-purged dimethylformamide (DMF) (1 mL), and CuOAc (9 mg, 73 μmol, 0.02 equiv) was added. The reaction mixture was placed in the microwave reactor (Biotage) and irradiated at 100 °C for 30 min. The clear solution was transformed into a turbid gel. The gel was dissolved in additional DMF (1 mL), 0.2 N HCl (20 mL) was added, and the aqueous solution was vortexed. The white precipitate was centrifuged, and the pellet

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10.1021/bm061010g CCC: $37.00
Published on Web 12/30/2006

Table 1. Reaction Conditions for the Polymerization of **1**

entry	reaction conditions	M_n[a]	M_w[b]	PDI[c]	T_g (°C)
1	200 mg **1**, $CuSO_4$/Na-ascorbate, RT, 3 days in 1 mL DMF/H_2O (95:5 v/v)	1100	1700	1.52	n.d.
2	500 mg **1**, $CuSO_4$/Na-ascorbate, RT, 3 days in 1 mL DMF/H_2O (95:5 v/v)	6900	12 700	1.74	n.d.
3	500 mg **1**, CuOAc, RT, 3 days in 1 mL DMF	27 600	55 000	1.99	n.d.
4	500 mg **1**, CuOAc, μW 100 °C, 30 min in 1 mL DMF	38 000	75 700	1.99	n.d.
5	1000 mg **1**, CuOAc, μW 100 °C, 30 min in 1 mL DMF	44 700	77 900	1.84	169
6	**1**, melt	46 700	86 300	1.84	165
7	**1**, melt, CuOAc	[d]	[d]	[d]	169
8	50 mg **1**, CuOAc, μW 100 °C, 30 min in 1 mL DMF	3060	8000	2.61	124
9	500 mg **1**, CuOAc, oil bath 100 °C, 30 min in 1 mL DMF	7050	8600	1.22	n.d.

[a] M_n (number average molecular mass) was determined by GPC with 10 mM LiCl in DMF, as eluens and PEG standards were used for calibration. [b] M_w (weight average molecular mass). [c] PDI: polydispersity index. [d] Polymerization product could not be analyzed due its insolubility in organic solvents used for spc; n.d.: not determined.

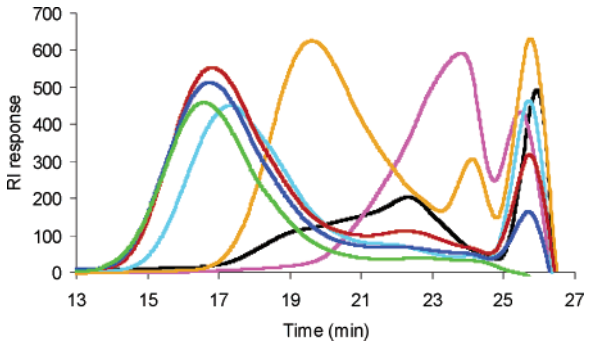

Figure 2. GPC chromatograms from the different polymerization reactions. Entry 1: (pink) up to linear tetramers; Entry 2: (orange) small polymers (6900); Entry 3: (turquoise) medium-sized polymers (27 600); Entry 4: (red) microwave heating: large polymers (38 000); Entry 5: (blue) microwave heating and increased concentration: large polymers (44 700); Entry 6: (green) polymerization in the melt (46 700), absence of copper (I); Entry 8: (black) cyclic oligomers.

was washed with 0.2 N HCl. The obtained solid was dissolved in AcOH and lyophilized, and the crude polymer was obtained in 92% yield (920 mg). ^{1}H NMR (dimethyl sulfoxide (DMSO)-d_6, 500 MHz) δ: 1.20 (m, 3H, βCH_3 Ala), 3.34/3.39 (double m, 2H, βCH_2 Phe), 4.28 (m, 3H, αCH Ala/CH_2), 5.55/5.70 (m, 1H, αCH Phe), 7.15 (m, 5H, arom H), 8.08 (s, 1H, C═C*H*, triazole), 8.41 (d, 1H, NH), 8.83 (d, 1H, NH), ^{13}C NMR (DMSO-d_6, 75 MHz) δ: 18.3 (βCH_3 Ala), 34.1 (CH_2), 37.5 (βCH_2 Phe), 48.3 (αCH Ala), 63.4 (αCH Phe), 122.0 (*C*═CH), 126.6, 128.1, 128.7, 136.0 (arom C), 144.2 (*C*═CH), 167.1 (C═O, Phe), 171.4 (C═O, Ala); differential scanning calorimetry (DSC) T_g: 169 °C.

Results and Discussion

The initial polymerization reaction was carried out with 200 mg of azido-acetylene functionalized dipeptide **1** in 1 mL of DMF/H_2O in the presence of $CuSO_4$/Na-ascorbate (entry 1, Table 1). After 3 days of stirring at room temperature, the isolated white precipitate consisted of only small oligomers (up to tetramers, i.e., eight amino acid residues, Figure 3) as was determined by gel permeation chromatography (GPC), relative to poly(ethylene glycol) (PEG)-based molecular weight standards (Figure 2), and liquid chromatography−mass spectrometry (LC−MS). Since FTIR still showed a sharp signal at υ 2100 cm^{-1} of the azide functionality, it was concluded that these were *acyclic* oligomers. Increasing the dipeptide monomer concentration to 500 mg in 1 mL of DMF/H_2O (entry 2) resulted in the

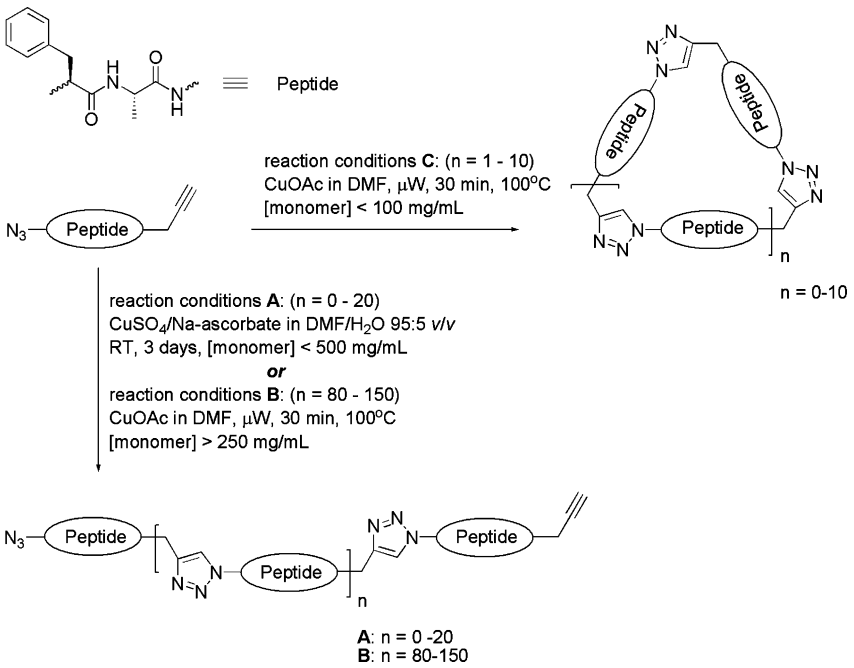

Figure 3. Structures of the synthesized peptide-based polymers. Reaction conditions **A** and **B** lead predominantly to linear polymerization products, while reaction condition **C** results in an increased ratio cyclic versus linear oligomer.

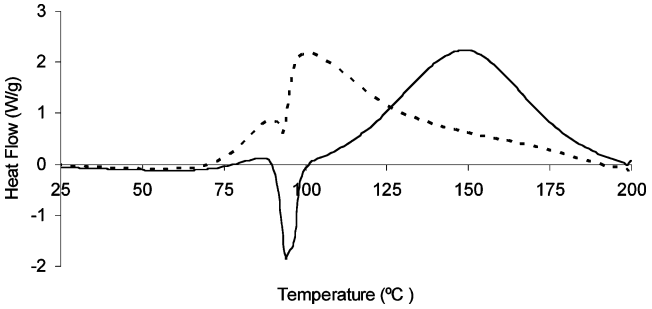

Figure 4. DSC thermograms of the polymerization of **1**. The polymerization was carried out in the melt; without CuOAc and with CuOAc (dotted line).

formation of a small polymer with M_n = 6900 Da (23-mer, 46 amino acid residues). Since the formation of copper (I) by the $CuSO_4$/Na-ascorbate redox couple could be the rate-limiting step of the click polymerization, copper (I) acetate was used instead. This led to a 4-fold increase in the molecular weight: M_n = 27 600 Da (92-mer, 184 amino acids, entry 3). Thus, besides increasing the azido-acetylene peptide monomer concentration, the presence of sufficient Cu(I) in the reaction mixture is a crucial parameter for increasing the molecular weight of the amino acid-based polymer.

Recently,[18] we showed the beneficial effect of heating by microwave irradiation upon 1,3-dipolar cycloaddition in order to attach a number of ligands to dendrimeric systems. Therefore, it was expected that further improvement of the polymerization reaction could be achieved in a microwave reactor. Indeed, the reaction of **1** at 500 and 1000 mg in 1 mL of DMF, respectively, with CuOAc as a catalyst in a microwave reactor at 100 °C (entries 4 and 5, respectively) gave already after 30 min a high molecular weight polymer, M_n = 38 000 and 44 700 Da, respectively (149-mer, ca. 300 amino acid residues, according to GPC analysis, Figure 2), as was apparent from the rapid transformation of the clear solution into a turbid gel. Microwave heating not only resulted in an increase of polymer length but also reduced the reaction time considerably. Moreover, the fact that the polydispersity index (M_w/M_n, Table 1) was near 2 was an additional indication for a high conversion rate of the polymerization reaction, according to the Flory equation[19] for typical step polymerizations.

A further, albeit slight, increase in M_n of the polymer (to ca. 46 700) was achieved by carrying out the polymerization in the melt in a DSC apparatus (Figure 4), since 1 g in 1 mL of DMF was close to saturation. Azido-acetylene peptide **1** was stable up to 250 °C, as verified by thermogravimetric analysis.

First, the polymerization in the melt was carried out in the absence of any copper (I) (entry 6), which clearly showed that the melting point of **1** was 91 °C. Increasing the temperature (10 °C/min) resulted in a sharp increase in the heat flow due to the exothermic polymerization of **1**. As was indicated above, a high molecular weight polymer, M_n = 46 700 Da (156-mer, >300 amino acid residues), was formed. However, as polymerization was carried out in the absence of copper (I), ^{1}H NMR analysis clearly showed the presence of both cycloaddition products (1,4-regioisomer: δ_H(triazole): 8.07 ppm, 1,5-regioisomer: δ_H(triazole): 7.41 ppm, both measured in DMSO-d_6). Next, in the presence of CuOAc (entry 7), the cycloaddition of **1** took place at a lower temperature, and the exothermic reaction was even more pronounced than it was in the absence of CuOAc (Figure 4). Unfortunately, this polymeric material could not be analyzed further due to its insolubility in organic solvents used for GPC analysis (e.g., DMF, tetrahydrofuran).

Thus, the microwave-assisted Cu(I)-catalyzed cycloaddition of **1** at a concentration of 1000 mg in 1 mL of DMF led to high molecular weight peptide-based polymers uniformly containing a 1,4-substituted triazole linking moiety.

Interestingly, when the microwave-assisted Cu(I)-catalyzed click polymerization was carried out at intermediate concentrations (50−250 mg of **1** per milliliter of solvent), GPC analysis of the isolated polymerization products always showed a bimodal molecular weight distribution (Figure 2, entry 8). It was anticipated that the low molecular weight part would contain both cyclic and linear oligomers (Figure 3, reaction conditions **C**). To this end, a sample that was polymerized at 50 mg/mL solvent (entry 8) was fractionated by preparative HPLC and analyzed by FTIR, MALDI-TOF, and LC−MS (Figure 5). Also, a second sample (entry 1, 200 mg **1** per milliliter of solvent, Cu(II), RT, reaction conditions **A**) was purified by HPLC and subsequently analyzed. In both samples, the linear and cyclic oligomers could be separated and characterized. FTIR *in combination with* mass spectrometry were used to distinguish

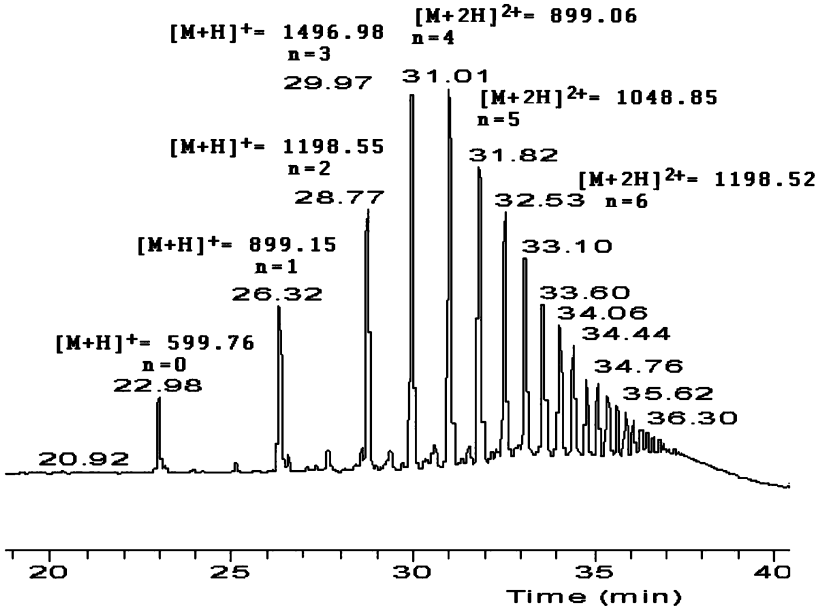

Figure 5. LC−MS pattern of polymerization product (entry 8) showing the cyclic oligomers (n = 0−6). Average Δ m/z = 299, corresponding to the mass of the dimer repeat unit.

between linear and cyclic peptide-based oligomers, since only the linear derivatives showed an intense peak at υ 2100 cm^{-1}, corresponding to the azide moiety. These HPLC analyses made it clear that the sample in entry 1 consisted primarily of short linear oligomers, while the sample represented by entry 8 consisted mainly of small cyclic oligomers. The smallest cyclic triazole oligomer that was identified contained two dipeptide units (four amino acid residues, $n = 0$ in Figures 3 and 5).[20] Although the ratio of cyclic versus linear oligomers could be influenced by choosing the reaction conditions, the exact value of this ratio could not be determined accurately. Generally, cyclic oligomers were predominantly formed at low monomer concentrations (50−250 mg **1** per milliliter of solvent, reaction conditions **C**), while, at higher concentrations of **1**, preferentially linear oligomers were obtained (reaction conditions **A** and **B**).

In a control experiment, monomer **1** (dissolved in 1 mL of DMF, entry 9) was subjected to a Cu(I)-catalyzed click polymerization, which was carried out at 100 °C by using a conventional oil bath. After 30 min, a sample was drawn and analyzed by GPC, while the remainder of the reaction mixture was kept at 100 °C. After 2 h and 24 h, another sample was drawn and analyzed by GPC. It was found that conventional heating did not result in high molecular weight polymers as compared to microwave heating (compare entry 9 with entry 4, Table 1). Increasing the reaction time up to 24 h was not effective since a significant increase in the molecular weight of the reaction products was not observed. Apparently, the click polymerization reaction needs very efficient heating, as is provided by a microwave apparatus in the first 30 min, to obtain high molecular weight polymers.

Conclusions

We have synthesized high molecular weight amino acid-based polymers in a backbone polymerization connecting their *N*- and *C*-termini employing the Cu(I)-catalyzed 1,3-dipolar cycloaddition reaction. To obtain high molecular weight polymers, the microwave heating was found to be superior to conventional heating. Moreover, the right choice of the reaction conditions was found to be an important factor to obtain either large linear polymers or medium-sized peptide macrocyclic oligomers. These optimized reaction conditions will be applied for the synthesis of biologically relevant peptide-based biopolymers.

Acknowledgment. Mies van Steenbergen (Department of Pharmaceutics) is acknowledged for his assistance with the GPC and (M)DSC analyses.

Supporting Information Available. Experimental procedures for the synthesis and characterization of **1** and characterization details (NMR, GPC, FTIR, DSC, MALDI-TOF, and LC−MS) of the polymers. This material is available free of charge via the Internet at http://pubs.acs.org.

References and Notes

(1) (a) Deming, T. J. *Adv. Mater.* **1997**, *9*, 299−311. (b) van Hest, J. C. M.; Tirrell, D. A. *Chem. Commun.* **2001**, 1897−1904. (c) Klok, H.-A. *Angew. Chem., Int. Ed.* **2002**, *41*, 1509−1513.

(2) (a) O'Brien-Simpson, N. M.; Ede, N. J.; Brown, L. E.; Swan, J.; Jackson, D. C. *J. Am. Chem. Soc.* **1997**, *119*, 1183−1188. (b) Sanda, F.; Endo, T. *Macromol. Chem. Phys.* **1999**, *200*, 2651−2661. (c) Ayres, L.; Adams, P. H. H. M.; Löwik, D. W. P. M.; van Hest, J. C. M. *Biomacromolecules* **2005**, *6*, 825−831.

(3) Jenkins, C. L.; Raines, R. T. *Nat. Prod. Rep.* **2002**, *19*, 49−59.

(4) Atkins, E. *Nature* **2003**, *424*, 1010.

(5) Tachibana, Y.; Fletcher, G. L.; Fujitani, N.; Tsuda, S.; Monde, K.; Nishimura, S.-I. *Angew. Chem., Int. Ed.* **2004**, *43*, 856−862.

(6) (a) Sever, M. J.; Weisser, J. T.; Monahan, J.; Srinivasan, S.; Wilker, J. J. *Angew. Chem., Int. Ed.* **2004**, *43*, 447−450. (b) Statz, A. R.; Meagher, R. J.; Barron, A. E.; Messersmith, P. B. *J. Am. Chem. Soc.* **2005**, *127*, 7972−7973.

(7) Crookes, W. J.; Ding, L.-L.; Huang, Q. L.; Kimbell, J. R.; Horwitz, J.; McFall-Ngai, M. J. *Science* **2004**, *303*, 235−238.

(8) Kricheldorf, H. R. *Alpha-aminoacid-N-carboxyanhydrides and Related Heterocycles*; Springer: Berlin, 1987.

(9) (a) Gallot, B. *Prog. Polym. Sci.* **1996**, *21*, 1035−1088. (b) Deming, T. J. *Nature* **1997**, *390*, 386−389. (c) Tsutsumiuchi, K.; Aoi, K.; Okada, M. *Macromolecules* **1997**, *30*, 4013−4017. (d) Schlaad, H.; Antonietti, M. *Eur. Phys. J. E* **2003**, *10*, 17−23. (e) Dimitrov, I.; Schlaad, H. *Chem. Commun.* **2003**, 2944−2945. (f) Koga, T.; Taguchi, K.; Kobuke, Y.; Kinoshita, T.; Higuchi, M. *Chem.—Eur. J.* **2003**, *9*, 1146−1156. (g) Sanda, F.; Gao, G.; Masuda, T. *Macromol. Biosci.* **2004**, *4*, 570−574. (h) Aliferis, T.; Iatrou, H.; Hadjichristidis, N. *Biomacromolecules* **2004**, *5*, 1653−1656. (i) Meyer, M.; Schlaad, H. *Macromolecules* **2006**, *39*, 3967−3970.

(10) Okamoto, K.; Rapaka, R. S.; Urry, D. W. *Biopolymers* **1978**, *17*, 573−591.

(11) Tachibana, Y.; Matsubara, N.; Nakajima, F.; Tsuda, T.; Tsuda, S.; Monde, K.; Nishimura, S.-I. *Tetrahedron* **2002**, *58*, 10213−10224.

(12) Metzke, M.; O'Connor, N.; Maiti, S.; Nelson, E.; Guan, Z. *Angew. Chem., Int. Ed.* **2005**, *44*, 6529−6533.

(13) (a) Tornøe, C. W.; Christensen, C.; Meldal, M. *J. Org. Chem.* **2002**, *67*, 3057−3064. (b) Rostovtsev, V. V.; Green, L. G.; Fokin, V. V.; Sharpless, K. B. *Angew. Chem., Int. Ed.* **2002**, *41*, 2596−2599.

(14) For reviews, see (a) Kolb, H. C.; Finn, M. G.; Sharpless, K. G. *Angew. Chem., Int. Ed.* **2001**, *40*, 2004−2021. (b) Breinbauer, R.; Köhn, M. *ChemBioChem* **2003**, *4*, 1147−1149. (c) Bock, V. D.; Hiemstra, H.; van Maarseveen, J. H. *Eur. J. Org. Chem.* **2006**, 51−68.

(15) For polymerization reactions, see (a) Opsteen, J. A.; van Hest, J. C. M. *Chem. Commun.* **2005**, 57−59. (b) Dirks, A. J.; van Berkel, S. S.; Hatzakis, N. S.; Opsteen, J. A.; van Delft, F. L.; Cornelissen, J. J. L. M.; Rowan, A. E.; van Hest, J. C. M.; Rutjes, F. P. J. T.; Nolte, R. J. M. *Chem. Commun.* **2005**, 4172−4174. (c) van Steenis, D. J. V. C.; David, O. R. P.; van Strijdonck, G. P. F.; van Maarseveen, J. H.; Reek, J. N. H. *Chem. Commun.* **2005**, 4333−4335. (d) Angelo, N. G.; Arora, P. S. *J. Am. Chem. Soc.* **2005**, *127*, 17134−17135. (e) Srinivasachari, S.; Liu, Y.; Zhang, G.; Prevette, L.; Reinecke, T. M. *J. Am. Chem. Soc.* **2006**, *128*, 8176−8184. (f) Bakbak, S.; Leech, P. J.; Carson, B. E.; Saxena, S.; King, W. P.; Bunz, U. H. F. *Macromolecules* **2006**, *39*, 6793−6795.

(16) Horne, W. S.; Yadav, M. Y.; Stout, C. D.; Ghadiri, M. R. *J. Am. Chem. Soc.* **2004**, *126*, 15366−15367.

(17) For recent applications in polymer synthesis/modification, see (a) Helms, B.; Mynar, J. L.; Hawker, C. J.; Fréchet, J. M. J. *J. Am. Chem. Soc.* **2004**, *126*, 15020−15021. (b) Tsarevsky, N. V.; Sumerlin, B. S.; Matyjaszewski, K. *Macromolecules* **2005**, *38*, 3558−3561. (c) Parrish, B.; Breitenkamp, R. B.; Emrick, T. *J. Am. Chem. Soc.* **2005**, *127*, 7404. (d) Laurent, B. A.; Grayson, S. M. *J. Am. Chem. Soc.* **2006**, *128*, 4238−4239. (e) Malkoch, M.; Thibault, R. J.; Drockenmuller, E.; Messerschmidt, M.; Voit, B.; Russell, T. P.; Hawker, C. J. *J. Am. Chem. Soc.* **2005**, *127*, 14942−14949. (f) Ladmiral, V.; Mantovani, G.; Clarkson, G. J.; Cauet, S.; Irwin, J. L.; Haddleton, D. M. *J. Am. Chem. Soc.* **2006**, *128*, 4823−4830. (g) Malkoch, M.; Vestberg, R.; Gupta, N.; Mespouille, L.; Dubois, P.; Mason, A. F.; Hedrick, J. L.; Liao, Q.; Frank, C. W.; Kingsbury, K.; Hawker, C. J. *Chem. Commun.* **2006**, 2774−2776.

(18) (a) Joosten, J. A. F.; Tholen, N. T. H.; Ait El Maate, F.; Brouwer, A. J.; van Esse, G. W.; Rijkers, D. T. S.; Liskamp, R. M. J.; Pieters, R. J. *Eur. J. Org. Chem.* **2005**, 3182−3185. (b) Rijkers, D. T. S.; van Esse, G. W.; Merkx, R.; Brouwer, A. J.; Jacobs, H. J. F.; Pieters, R. J.; Liskamp, R. M. J. *Chem. Commun.* **2005**, 4581−4583.

(19) Flory, P. J. *Principles of Polymer Chemistry*; Cornell University Press: Ithaca, NY, 1953; Chapter 2.

(20) (a) Punna, S.; Kuzelka, J.; Wang, Q.; Finn, M. G. *Angew. Chem., Int. Ed.* **2005**, *44*, 2215−2220. (b) van Maarseveen, J. H.; Horne, W. S.; Ghadiri, M. R. *Org. Lett.* **2005**, *7*, 4503−4506. (c) Angell, Y.; Burgess, K. *J. Org. Chem.* **2005**, *70*, 9595−9598. (d) Bock, V. D.; Perciaccante, R.; Jansen, T. P.; Hiemstra, H.; van Maarseveen, J. H. *Org. Lett.* **2006**, *8*, 919−922.

BM061010G