Synthesis and Solution Properties of Norbornene Based Polybetaines

Semra Colak and Gregory N. Tew*

Department of Polymer Science and Engineering, University of Massachusetts-Amherst, Conte Research Center for Polymers, 120 Governor's Drive, Amherst, Massachusetts 01003

Received June 9, 2008; Revised Manuscript Received September 11, 2008

ABSTRACT: Ring-opening metathesis polymerization (ROMP) was used to synthesize novel norbornene based polycarboxy- (**d-Poly 3a** and **d-Poly 3b**) and polysulfobetaines (**Poly 3c**) using the third generation Grubbs' catalyst (G3) as the initiator. Hydrophobicity of the polycarboxybetaines was varied by changing the bridging group of the norbornene backbone. A protective group approach was utilized to prevent any possible retardation in the polymerization due to interactions of the carboxylate functionality with the catalyst and to provide ease of characterization. The *tert*-butyl ester protected precursor polymers (**Poly 3a** and **Poly 3b**) were deprotected under acidic conditions to yield the corresponding polycarboxybetaines with very narrow polydispersity indices, ranging from 1.03 to 1.15. This method allowed excellent control over the molecular weight distributions compared to the direct polymerization approach. When molecular weight was plotted against the theoretical degree of polymerization (DP), linear relationships were obtained for both ^{1}H NMR and GPC-MALLS data. Oxanorbornene based polycarboxybetaine (**d-Poly 3a**) was studied in aqueous 0.1 M NaBr solution by dynamic light scattering (DLS) and no significant aggregation was observed. An attempt was made to determine the acid ionization constant (K_a) of the carboxylate group, which led to the discovery that the cyclic imide of these particular monomers is easily ring opened under basic conditions.

Introduction

Polymeric betaines constitute a special class of zwitterionic polymers. They have the positive and negative charges on the same repeat unit, ensuring net neutral charge on the polymer, unlike polyampholytes, which are made from both cationic and anionic monomers.[1,2] Depending on the nature of the ionizable (anionic) group, polybetaines are categorized as carbo-,[3−5] sulfo[6,7] and phosphobetaines[8] and typically contain quaternary ammoniums as the cationic group. They have many important solution properties, the most interesting of which is their insolubility in pure water due to formation of intra- and interchain associations. However, they become soluble in water upon addition of salt (e.g., NaCl), which screens the electrostatic associations. This is known as the antipolyelectrolyte effect.[2,9] Polymeric betaines have found many applications in biotechnology, hydrometallurgy, oil industries, and medicine.[10−12] Recently, with the developments in controlled living polymerization techniques, they have gained more interest for their bio- and hemocompatible properties. When incorporated into materials or used as surface coatings, some zwitterionic polymers were found to strongly resist protein absorption, due to their highly hygroscopic nature.[13−16] Therefore, these materials are promising candidates for applications in the fields of medical diagnostics, drug delivery, and tissue engineering.

Synthetic routes to polybetaines have been either via direct polymerization of a zwitterionic monomer or post functionalization of a precursor polymer. The most commonly employed polymerization technique has been conventional free radical polymerization as demonstrated by Salamone et al.[9,17] and Laschewsky and co-workers.[4,18] Even though there has been significant interest in the synthesis and applications of new polymeric betaines, there are few examples of well-defined systems via controlled living polymerization techniques. Early examples were reported by Armes and co-workers, where they demonstrated the polymerization of 2-(dimethylamino)ethyl methacrylate via group transfer polymerization (GTP), which was then betainized with 1,3-propanesultone to yield polybetaines with narrow molecular weight distributions.[19,20] Later examples include polybetaines synthesized via reversible addition-fragmentation chain transfer polymerization (RAFT)[21−23] and atom transfer radical polymerization (ATRP).[24,25] Another living polymerization technique, which has attracted great attention in the last two decades following improvements in the homogeneous catalyst systems, is ring-opening metathesis polymerization (ROMP). Even though many types of catalyst systems have been developed, Ruthenium based ones are the most successful due to their higher reactivity rates, prolonged stability, and functional group tolerance.[26−28] There were no reports on polybetaines via ROMP until very recently when Lowe and co-workers[29] demonstrated the polymerization of oxanorbornene based betaines using Grubbs' first generation catalyst. In their paper, sulfopropylbetaine-exo-7-oxanorbornene was polymerized directly, whereas the carboxylate functionality of carboxyethylbetaine-exo-7-oxanorbornene was protonated prior to polymerization.

In this report, we describe the synthesis of norbornene based polycarboxy- and polysulfobetaines via ROMP with the third generation Grubbs' catalyst as the initiator. ROMP is known to be tolerant to many functional groups; however, carboxylates have a retardation effect on the polymerization kinetics.[27] To avoid this effect and also to facilitate characterization, the synthesis of the polycarboxybetaine was carried out utilizing a protecting group method. The zwitterionic functionality was attained upon deprotection of the *tert*-butyl ester polymer under mild acidic conditions to yield the desired polycarboxybetaine. A direct polymerization approach was used for the synthesis of the sulfobetainic monomer. In addition to differentiating the ionic group, we varied the amphiphilicity of polycarboxybetaines by inserting a hydrophobic moiety at the bridging group. The solution properties of the polycarboxybetaine, **d-Poly 3a**, were analyzed with aqueous gel permeation chromatography multi-angle laser light scattering (GPC-MALLS) and dynamic light scattering (DLS) methods.

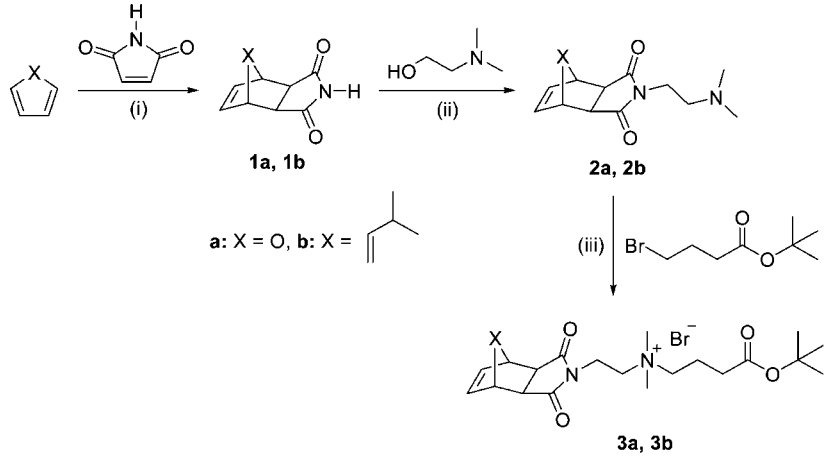
Results and Discussion

There are many examples demonstrating that ROMP can be successfully used to polymerize norbornene derivatives having

* To whom correspondence should be addressed. E-mail: tew@mail.pse.umass.edu.

10.1021/ma801295u CCC: $40.75
Published on Web 10/23/2008

Scheme 1. Synthesis of Monomers 3a and 3b[a]



[a] Conditions and reagents: (i) **1a**: ethyl acetate, at 90 °C for 3 h; **1b**: toluene, at 135 °C for 12 h. (ii) **2a** and **2b**: THF, Ph_3P, DIAD, at RT for 24 h. (iii) **3a** and **3b**: THF, at 50 °C for 36 h.

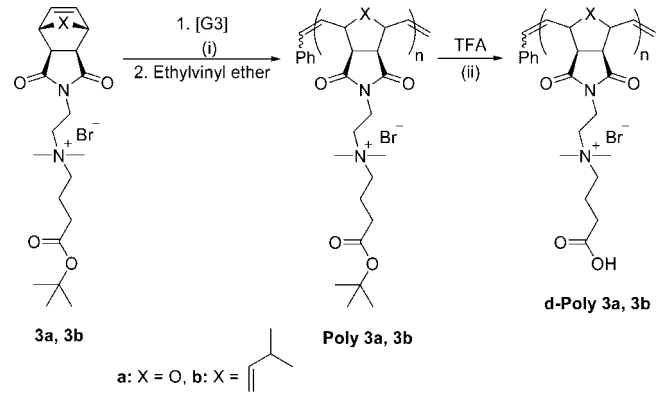
different functional groups.[30−34] However, it was also reported that the presence of particular additives such as pyridine, secondary amines, thiols, or benzoic acid, can significantly slow or shut down the ROMP kinetics.[27] Therefore, to prevent this retardation effect and to synthesize norbornene based polycarboxybetaines via ROMP, a protecting group approach was utilized. The carboxylate groups of monomers **3a** and **3b** were protected with *tert*-butyl groups that could be removed under acidic conditions after polymerization, to yield the desired polycarboxybetaine.

Monomers were synthesized via a three step procedure (Scheme 1). A Diels−Alder reaction of the corresponding diene with maleimide yielded **1a** and **1b**. To demonstrate that polycarboxybetaines having different amphiphilicity can be synthesized via ROMP, two different diene derivatives were used. Furan was used to yield a relatively more hydrophilic 7-oxanorbornene backbone, whereas isopropylfulvene provided a more hydrophobic backbone. The products were purified to obtain pure exo isomers, which are known to be much more reactive than the corresponding endo isomers under ROMP conditions.[35] In the next step, the imide groups of **1a** and **1b** were coupled with *N*,*N*-dimethylethanolamine via the Mitsunobu reaction to yield the precursor tertiary amines **2a** and **2b**, which were then alkylated with 4-bromobutanoic acid *tert*-butyl ester to yield the protected monomers **3a** and **3b** (Scheme 1). The sulfobetaine analog was synthesized using similar procedures (see Supporting Information).

Depending on the ionic functionality and the hydrophobicity of the monomers, different solvent systems were required to provide a homogeneous medium for the polymerization reactions. The third generation Grubbs' catalyst was used as the initiator for all the polymerizations and they were terminated using ethyl vinyl ether. Monomer **3a** was homopolymerized using only DCM, whereas **3b** required a mixture of THF and methanol (Scheme 2). After polymerization, the resulting homopolymers **Poly 3a** and **Poly 3b** were subjected to deprotection to yield the corresponding polycarboxybetaines **d-Poly 3a** and **d-Poly 3b** (Scheme 2). ^{1}H NMR was used to characterize the extent of deprotection (Figure 1) and it was clearly observed that no residual *tert*-butyl ester protons were present after reacting with neat TFA, indicating complete deprotection. Because **d-Poly3b** was visibly insoluble in aqueous solutions, it was not studied further in this report.

Certain conditions are required for a polymerization to be considered living and controlled.[36] Provided that the system is living, a linear relationship should be obtained between DP and

Scheme 2. Polymerization Procedure for 3a and 3b and Deprotection of Poly 3a and Poly 3b[a]



[a] Reagents and conditions: (i) **Poly 3a**: DCM, at RT for 20 min; **Poly 3b**: THF, methanol, at 60 °C for 1 h. (ii) **d-Poly 3a, 3b**: at RT for 24 h.

the molecular weight. In order to analyze the living behavior of ROMP for this particular system, a series of molecular weights of **Poly 3a** were synthesized (Table 1). The DPs were calculated by taking the ratio of the monomer concentration to the initiator concentration. The experimental molecular weights were determined using two different methods, ^{1}H NMR and aqueous gel permeation chromatography coupled with a multi angle laser light scattering (GPC-MALLS) detector. The molecular weight of the protected polymers, **Poly 3a**, were determined from the ^{1}H NMR data by comparing the integration values of the phenyl end group protons to the tert-butyl protons in the monomer repeat unit. The resulting experimental data were in accordance with the theoretical values (Table 1).

These polymers were then deprotected to obtain the **d-Poly 3a** samples, which were analyzed using GPC-MALLS, yielding *absolute molecular weights*. GPC of charged polymers is challenging due to ionic aggregations or interactions with the stationary phase of the chromatographic column.[37] Careful sample preparation is needed in order to obtain accurate data. A high salt concentration aqueous solution, 0.1 M NaBr, was used as the eluent and the samples were stirred overnight to dissolve any possible aggregates. To eliminate interactions of the polymer samples with the packing material, a Suprema MAX column was used. Monomodal, symmetrical peaks with narrow molecular weight distributions were obtained for all the tested

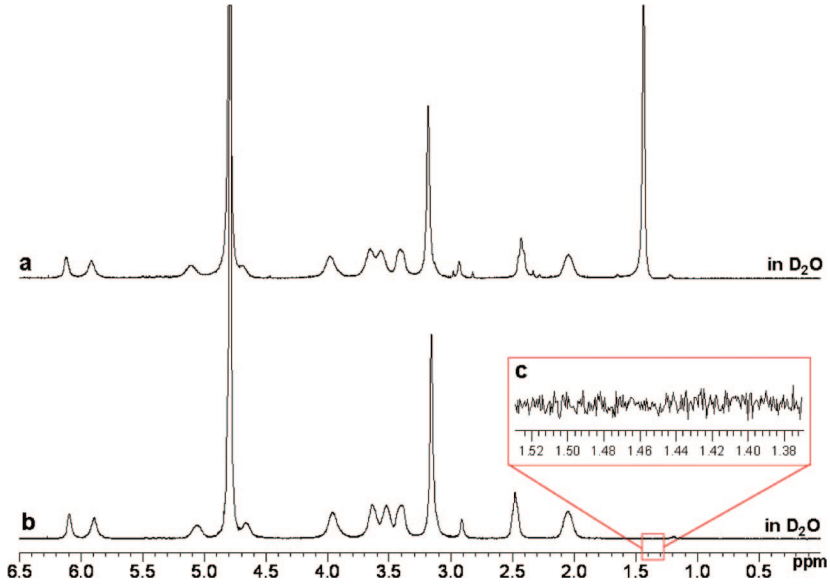


Figure 1. Deprotection of **Poly 3a**. (a) ^{1}H NMR of **Poly 3a** (protected), (b) **d-Poly 3a** (deprotected), and (c) inset highlights that, within the resolution of ^{1}H NMR, the deprotection is complete.

Table 1. Molecular Weight Characterization of Poly 3a and d-Poly 3a

	$M_{n\ (theoretical)}$ [kDa]		$M_{n\ (experimental)}$ [kDa]				
			^{1}H NMR		GPC-MALLS[b,c]		
[M]/[I]	$M_{n\ (protected)}$	$M_{n\ (deprotected)}$	M_n[a]	M_n[b]	M_n	M_w[c]	PDI[b]
6.6	3.0	2.7	3.3	3.0	n.d.	n.d.	n.d.
12.4	5.7	5.0	6.9	5.5	11.3	13.1	1.15
17.5	8.0	7.0	8.1	7.9	12.3	14.2	1.15
24.9	11.4	10.0	12.8	12.4	18.3	20.3	1.10
54.6	25.1	22.0	28.1	24.9	37.3	41.9	1.12
83.4	38.3	33.6	40.5	37.3	52.7	54.2	1.03
125.9	57.8	50.8	63.1	50.8	75.6	79.9	1.06
251.9	115.7	101.6	n.d.	n.d.	144.7	152.1	1.05

[a] Results for **Poly 3a** (protected). [b] Results for **d-Poly 3a** (deprotected). [c] The specific refractive index increment (dn/dc) was determined to be 0.149 ± 0.002 mL/g.

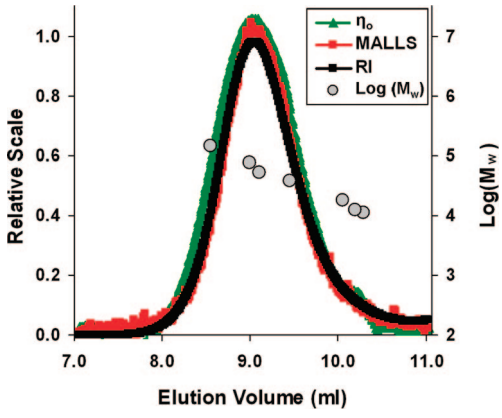


Figure 2. GPC-MALLS trace of **d-Poly 3a**, $M_{w\ (GPC\text{-}MALLS)}$ = 54.2 kDa.

samples and Figure 2 shows a representative chromatogram of a **d-Poly 3a** sample (54.2 kDa), including a second axis showing the elution volumes of all the samples with varying molecular weights from the lowest at 10.3 mL to the highest at 8.5 mL. The experimental molecular weights obtained from the GPC-MALLS analysis, reported in Table 1, were relatively higher than the theoretical values. On the other hand, the molecular weights calculated by ^{1}H NMR agreed quite well with the

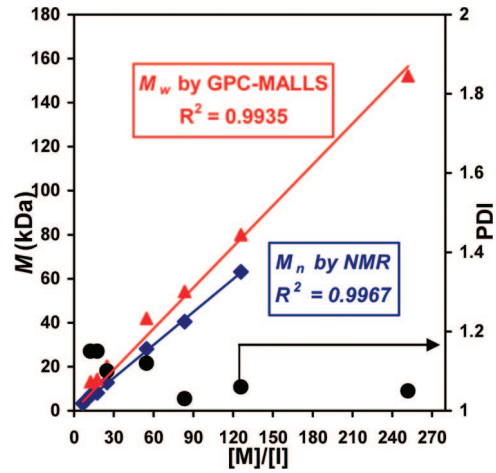


Figure 3. Molecular weight dependence of **Poly 3a** by ^{1}H NMR (triangles) and **d-Poly 3a** by GPC-MALLS (squares) on [M]/[I]. PDI (solid circles) was determined from the GPC-MALLS data.

theoretical values, and it is well known that molecular weight determination by light scattering for charged systems is complicated.[38]

Figure 3 shows the plot of the experimental molecular weights determined by ^{1}H NMR and GPC-MALLS versus theoretical degree of polymerization (DP) values (ratio of the monomer concentration to the initiator concentration). It is clearly seen that linear relationships were obtained for both methods, which proves the living behavior of this system.

Dynamic light scattering (DLS) is one of the best techniques to determine whether a polymer is soluble in a particular solvent system or not. If soluble, polymer chains should exist as single chains with no aggregate formation. As mentioned before, zwitterionic polymers are insoluble in water but become soluble provided there is a high enough salt concentration in the media. DLS was performed to analyze if the salt concentration in the GPC-MALLS experiments was indeed adequate, or not, to prevent large scale aggregate formation. Samples of **d-Poly 3a** with an absolute molecular weight of 79.9 kDa were prepared at varying concentrations (from 0.5 to 10.0 g/L) in 0.1 M NaBr solution, which was used in the GPC-MALLS experiment. Figure 4a shows the field correlation function and relaxation

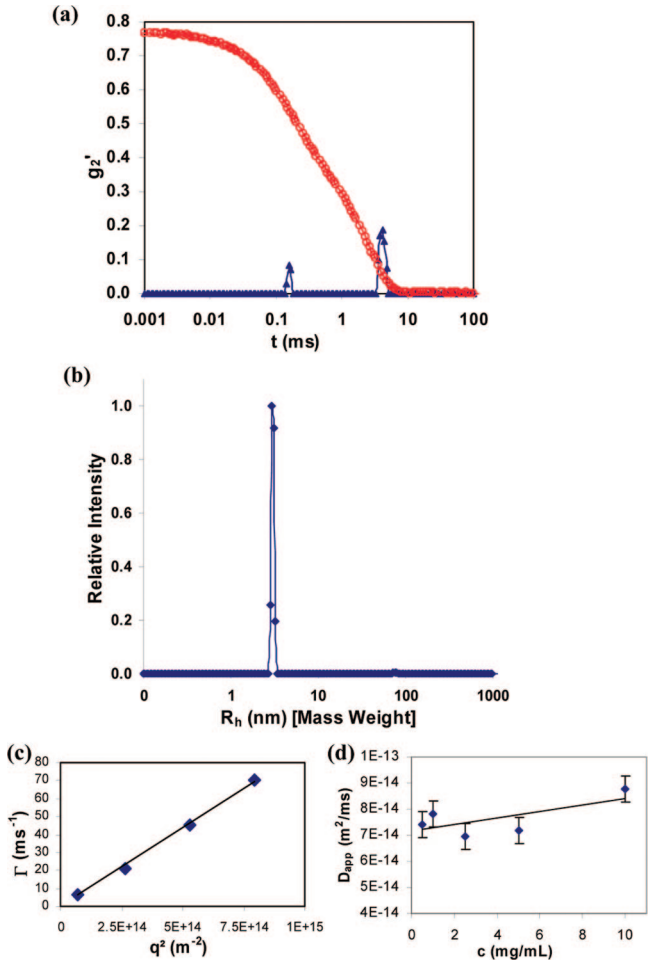


Figure 4. Dynamic light scattering data of **d-Poly 3a**, $M_{w\ (GPC\text{-}MALLS)}$ = 79.9 kDa, (a) field correlation function (red open circles) and relaxation time distribution (blue triangles), (b) hydrodynamic radius distribution (blue diamonds), (c) plot of Γ vs q^2 for a representative sample, and (d) determination of diffusion coefficient at different concentrations (R_h = 3.4 nm).

time distributions, while Figure 4b shows the mass weighted radius distributions of the corresponding relaxation times.

Interestingly, even though two distinct relaxation times were observed in the field correlation function, when converted to mass weighted average an intense peak with a very narrow distribution dominated the graph along with a much smaller weakly intense peak (Figure 4b). This indicates that there are few aggregates of negligible population compared to the population of single chains in this solvent. The corresponding hydrodynamic radius (R_h) of the main peak was determined to be 3.4 nm by double extrapolation to zero scattering angle and zero concentration (Figure 4c and d), which is consistent with the size of this single chain. Using the same approach, the hydrodynamic radius of the aggregates was determined to be 73.8 nm.

Depending on the pH of the solution or whether it is below or above its isoelectric point, a polybetaine of this type can be overall zwitterionic or cationic. An attempt was made to determine the acid ionization constant of **d-Poly3a**. For that purpose, the monomer **3a** was deprotected initially with the same procedure used for the deprotection of **Poly 3a**. It was dissolved in neat TFA and stirred overnight, followed by precipitation in excess diethyl ether. The titration was done with a 15 mL of 10.0 mg/mL solution of the deprotected monomer and a standard 0.1 M NaOH solution. A small amount of salt was added to the solution to provide the media with necessary conductance for the titration. As the titration approached neutral pH, it was observed that the system became unstable (pH varied randomly), therefore, data collection was terminated. To investigate the cause of this observation, the deprotected monomer was kept overnight in the same NaOH solution used for the titration. The sample was freeze-dried and characterized by ^{1}H NMR without any further purification. Figure 5 compares the ^{1}H NMR spectra of the deprotected monomer before and after the NaOH treatment.

When Figure 5a is compared to Figure 5b, it is clear that a new species is present. A possible explanation was the ring opening of the imide group under nucleophilic basic conditions. To completely characterize the product, both spectra were compared to a model compound (Figure 5c). This comparison

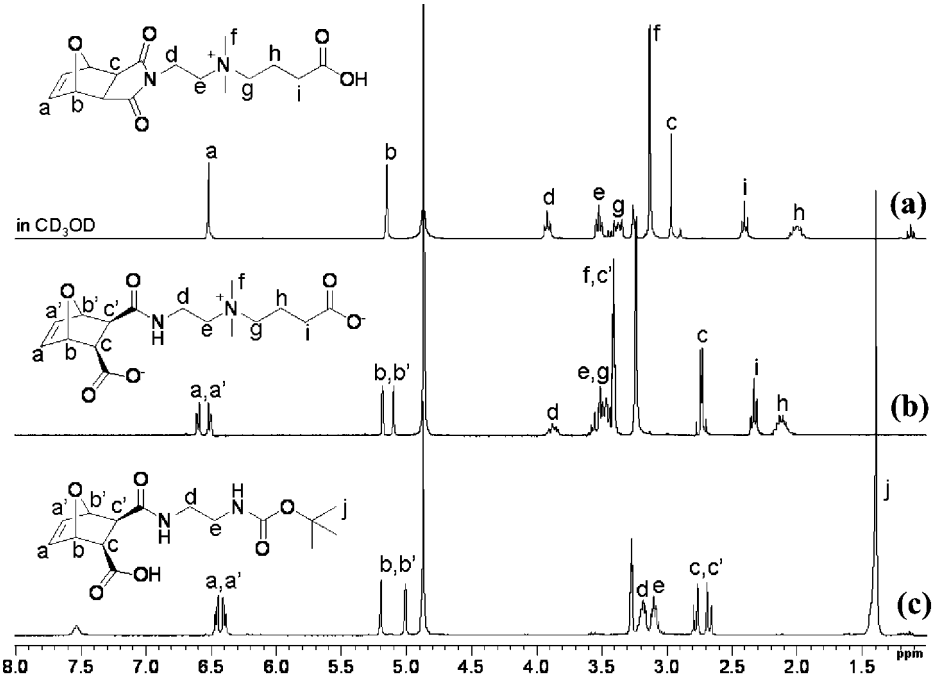


Figure 5. Stability of **3a** in basic nucleophilic media monitored by ^{1}H NMR. (a) ^{1}H NMR of monomer **3a** after deprotection, (b) ^{1}H NMR of deprotected **3a** after NaOH treatment, (c) ^{1}H NMR of a model compound.

confirmed that the splitting of the norbornene protons is due to ring opening of the imide group. A similar example was reported, where the imide stability of phthalimides was investigated under physiological conditions and demonstrated that *N*-(3-trimethylaminopropyl)phthalimido bromide readily undergoes hydroxide ion catalyzed hydrolysis.[39] It should be noted that this was an aromatic imide (phthalimide) as opposed to our aliphatic one. In addition, the imides reported here are stable to pH = 7.4.

Conclusions

Polybetaines with different hydrophobicities and ionizable groups were synthesized via ring-opening metathesis polymerization (ROMP), which resulted in well defined, living systems with narrow molecular weight distributions (PDI ranging from 1.03 to 1.15). When compared to the previously reported direct polymerization approach, utilizing a protecting group allowed us to have high control over the polydispersity of the polymers. Extensive characterization of the solution properties of **d-Poly 3a** by GPC-MALLS and DLS, proved that 0.1 M NaBr provided the necessary medium to break large scale aggregates.

With the ease and versatility of ROMP to synthesize more complex architectures or cross-linked networks, these polymers can be further incorporated into or used to synthesize materials. Keeping in mind that carboxylate and sulfonate containing polybetaines have very good bio- and hemocompatibilities, these materials appear to be good candidates for applications as drug delivery agents, nonfouling coatings, hydrogels, and tissue engineering scaffolds.

Acknowledgment. This work was supported by ONR (N00014-07-1-0520). Dr. Karen Lienkamp and Dr. Ahmad Madkour are gratefully acknowledged for their helpful discussions. Mass spectral data were obtained at the University of Massachusetts Mass Spectrometry Facility, which is supported, in part, by the National Science Foundation.

Supporting Information Available: All experimental procedures, including monomer and polymer synthesis, as well as detailed information about characterization techniques. This material is available free of charge via the Internet at http://pubs.acs.org.

References and Notes

(1) Kudaibergenov, S.; Jaeger, W.; Laschewsky, A. *Adv. Polym. Sci.* **2006**, *201*, 157–224.
(2) Lowe, A. B.; McCormick, C. L. *Chem. Rev.* **2002**, *102*, 4177–4189.
(3) Noh, J.-G.; Sung, Y.-J.; Geckeler, K. E.; Kudaibergenov, S. E. *Polymer* **2005**, *46*, 2183–2190.
(4) Bonte, N.; Laschewsky, A. *Polymer* **1996**, *37*, 2011–2019.
(5) Hamaide, T.; Germanaud, L.; Le Perchec, P. *Makromol. Chem.* **1986**, *187*, 1097–1107.
(6) Liaw, D.-J.; Huang, C.-C. *Macromol. Chem. Phys.* **2000**, *201*, 1101–1107.
(7) Kudaibergenov, S. E., Ed.; *Polyampholytes: Synthesis, Characterization and Application*; Kluwer Academic/Plenum Publishers: Dordrecht, The Netherlands, 2002.
(8) Nakaya, T.; Li, Y. J. *Prog. Polym. Sci.* **1999**, *24*, 143–181.
(9) Salamone, J. C.; Volksen, W.; Olson, A. P.; Israel, S. C. *Polymer* **1978**, *19*, 1157–1162.
(10) Koba, J.; Fukazawa, J.; Yoda, Y.; Kitsuki, T. JP 07223920, **1995**.
(11) Hellsten, M.; Harwigsson, I. WO 9628527, **1996**.
(12) Messadek, J. WO 2006050585, **2006**.
(13) Zhang, Z.; Chen, S.; Jiang, S. *Biomacromolecules* **2006**, *7*, 3311–3315.
(14) Chen, S.; Jiang, S. *Adv. Mater.* **2008**, *20*, 335–338.
(15) Zhang, Z.; Chao, T.; Chen, S.; Jiang, S. *Langmuir* **2006**, *22*, 10072–10077.
(16) Chang, Y.; Chen, S.; Zhang, Z.; Jiang, S. *Langmuir* **2006**, *22*, 2222–2226.
(17) Salamone, J. C.; Volksen, W.; Israel, S. C.; Olson, A. P.; Raia, D. C. *Polymer* **1977**, *18*, 1058–1062.
(18) Favresse, P.; Laschewsky, A. *Polymer* **2001**, *42*, 2755–2766.
(19) Lowe, A. B.; Billingham, N. C.; Armes, S. P. *Chem. Commun.* **1996**, *13*, 1555–1556.
(20) Lowe, A. B.; Billingham, N. C.; Armes, S. P. *Macromolecules* **1999**, *32*, 2141–2148.
(21) Donovan, M. S.; Sumerlin, B. S.; Lowe, A. B.; McCormick, C. L. *Macromolecules* **2002**, *35*, 8663–8666.
(22) Mertoglu, M.; Laschewsky, A.; Skrabania, K.; Wieland, C. *Macromolecules* **2005**, *38*, 3601–3614.
(23) Mertoglu, M.; Garnier, S.; Laschewsky, A.; Skrabania, K.; Storsberg, J. *Polymer* **2005**, *46*, 7726–7740.
(24) Lobb, E. J.; Ma, I.; Billingham, N. C.; Armes, S. P.; Lewis, A. L. *J. Am. Chem. Soc.* **2001**, *123*, 7913–7914.
(25) Li, C.; Tang, Y.; Armes, S. P.; Morris, C. J.; Rose, S. F.; Lloyd, A. W.; Lewis, A. L. *Biomacromolecules* **2005**, *6*, 994–999.
(26) Trnka, T. M.; Grubbs, R. H. *Acc. Chem. Res.* **2001**, *34*, 18–29.
(27) Slugovc, C. *Macromol. Rapid Commun.* **2004**, *25*, 1283–1297.
(28) Bielawski, C. W.; Grubbs, R. H. *Prog. Polym. Sci.* **2007**, *32*, 1–29.
(29) Rankin, D. A.; Lowe, A. B. *Macromolecules* **2008**, *41*, 614–622.
(30) Ilker, M. F.; Schule, H.; Coughlin, E. B. *Macromolecules* **2004**, *37*, 694–700.
(31) Ilker, M. F.; Nuesslein, K.; Tew, G. N.; Coughlin, E. B. *J. Am. Chem. Soc.* **2004**, *126*, 15870–15875.
(32) Eren, T.; Som, A.; Rennie, J. R.; Nelson, C. F.; Urgina, Y.; Nusslein, K.; Coughlin, E. B.; Tew, G. N. *Macromol. Chem. Phys.* **2008**, *209*, 516–524.
(33) Alfred, S. F.; Al-Badri, Z. M.; Madkour, A. E.; Lienkamp, K.; Tew, G. N. *J. Polym. Sci., Part A: Polym. Chem.* **2008**, *46*, 2640–2648.
(34) Rankin, D. A.; P'Pool, S. J.; Schanz, H.-J.; Lowe, A. B. *J. Polym. Sci., Part A: Polym. Chem.* **2007**, *45*, 2113–2128.
(35) Lapinte, V.; Brosse, J.-C.; Fontaine, L. *Macromol. Chem. Phys.* **2004**, *205*, 824–833.
(36) Webster, O. W. *Science* **1991**, *251*, 887–893.
(37) Volet, G.; Lesec, J. *J. Liq. Chromatogr.* **1994**, *17*, 559–577.
(38) Ruehe, J.; Ballauff, M.; Biesalski, M.; Dziezok, P.; Groehn, F.; Johannsmann, D.; Houbenov, N.; Hugenberg, N.; Konradi, R.; Minko, S.; Motornov, M.; Netz, R. R.; Schmidt, M.; Seidel, C.; Stamm, M.; Stephan, T.; Usov, D.; Zhang, H. *Adv. Polym. Sci.* **2004**, *165*, 79–150.
(39) Su, S. C. K.; Shafer, J. A. *J. Org. Chem.* **1969**, *34*, 2911–15.

MA801295U